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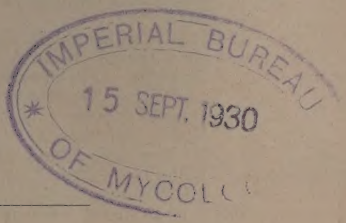
THE RHIZOCTONIA DAMPING-OFF OF CONIFERS, AND ITS CONTROL BY CHEMICAL TREATMENT OF THE SOIL

JAMES STEWART WIANT



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THE RHIZOCTONIA DAMPING-OFF OF CONIFERS, AND ITS CONTROL BY CHEMICAL TREATMENT OF THE SOIL[1]

JAMES STEWART WIANT

In 1924 the writer began a study of the fungicidal properties of certain organic mercury compounds in comparison with various inorganic chemicals. Early experiments in greenhouses demonstrated that several of these materials were effective in controlling damping-off of coniferous seedlings. Field tests were then carried out in a small experimental nursery at Ithaca, New York, during the season of 1925. The results obtained suggested the advisability of continuing the investigations on a more extensive scale. An attempt was therefore made to determine the importance of damping-off in a number of the larger nurseries in the northeastern United States and the neighboring provinces of Canada. It was found that damping-off was a serious problem in seven of the eleven nurseries from which replies were received. In none of these seven nurseries, and in but one of the other four, were treatments with chemicals being systematically employed for controlling the disease. The need for further work in eastern nurseries was therefore apparent, and arrangements were made to establish a field laboratory at the nursery of the Keene Forestry Association, Keene, New Hampshire, where investigations were carried on during the seasons of 1926 and 1927.

Since Rhizoctonia was almost the only pathogene involved in these experiments, the larger part of the work was done on the particular damping-off disease caused by this fungus.

HISTORY

This review aims to note the more important work on the damping-off of conifers, and to call special attention to those papers having a direct relation to the present investigations. A more detailed review of the literature prior to 1921 may be found in the excellent paper of Hartley (1921).

The problem of damping-off early engaged the attention of German foresters and nurserymen, who first attempted the mass production of coniferous seedlings. Much of their work was observational in character.

[1] Also presented to the Faculty of the Graduate School of Cornell University, June, 1928, as a major thesis in partial fulfillment of the requirements for the degree of doctor of philosophy.

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With the more recent development of reforestation projects and the consequent establishment of large coniferous nurseries in the United States, there has been a corresponding development of knowledge concerning this disease. Modern investigations have centered largely about the problems of etiology and control.

As pointed out by Hartley (1921:7), the only fungi definitely connected with the disease as it is discussed in the older European literature appear to be *Fusarium* spp. (*Fusoma* spp.) and *Phytophthora fagi* R. Hartig (*P. omnivora* De Bary in part). As far as the writer knows, *P. fagi* has not been reported from conifers in the United States.

Early reports of Fusarium damping-off in this country are those of Jones (1908), Gifford (1911), and Spaulding (1914). The last-named author finds forty-four different strains[2] capable of attacking pines. Hartley, Merrill, and Rhoads (1918) summarize the previous inoculation tests with Fusarium, and record the results of their own experiments with four common species. They agree with Spaulding that more than one species are involved and that *F. moniliforme* Sheldon (*Gibberella moniliformis* [Sheldon] Wineland) is one of the more virulent. More recently, Lindfors (1922) has isolated six species of Fusarium from pine in Sweden. His data indicate that these and two other species tested are pathogenic on conifers, but that four of the eight are decidedly more virulent than the others. Dounin and Goldmacher (1926) call attention to the prevalent opinion in Russia that but one species, *F. blasticola* Rostrup, is involved. From their tests they conclude that four additional species are pathogenic, but the data which they present are limited. It is of interest to note that these investigators claim to have isolated and studied a species of bacteria capable of producing damping-off in the absence of fungi. No details of their inoculation tests are given. Sibilia (1925), in Italy, has described several species of Fusarium from conifers, but evidently has not conducted inoculation tests.

Accounts of the relation of *Pythium debaryanum* Hesse to damping-off of conifers are given by Hartley (1910), Hofmann (1912), Spaulding (1914), and Hartley and Hahn (1919). In a later paper, Hartley (1921) reports extensive experiments which definitely prove the pathogenicity of this organism on pines. He demonstrates also that strains differ in their virulence, and shows that on the basis of morphology and pathogenicity the conifer strains as a group are not different from those attacking angiosperms.

Duggar and Stewart (1901) are evidently the first to have recorded the association of Rhizoctonia with the disease on conifers. Later reports are made by Hartley (1910) from Nebraska, and by Clinton (1913) from Connecticut. Hartley, Merrill, and Rhoads (1918), and Hartley (1921),

[2] The writer uses the term *strain* throughout this publication to indicate the fungus obtained in pure culture from a single isolation.

conclusively demonstrate the pathogenicity of Rhizoctonia on conifers, and on the basis of inoculation tests point out marked differences in virulence between the strains used. Together with the work of Edson (1915) with sugar beets, and that of Edson and Shapovalov (1918) with potatoes, the results indicate that strains pathogenic on conifers are likewise capable of attacking angiosperms, and vice versa. Hartley (1921) concludes therefore that the strains from conifers are identical with the common *Rhizoctonia solani* Kühn, the perfect stage of which is *Corticium vagum* B. & C. (Burt, 1926).

It is apparent from the literature (Duggar, 1915; Peltier, 1916; Rosenbaum and Shapovalov, 1917; Edson and Shapovalov, 1918; Matsumoto, 1921; Briton-Jones, 1924, 1925; Gratz, 1925; K. S. Thomas, 1925; Van der Meer, 1926) that marked differences exist between various strains of this common pathogene. The evidence presented by Rosenbaum and Shapovalov (1917), Matsumoto (1921), Gratz (1925), and Van der Meer (1926), suggests the existence of special forms within the species. Duggar (1915) and Matsumoto (1921) each report a single fungus which differs sufficiently from the others studied to possibly merit specific rank. Matz (1921), working with strains of Rhizoctonia found on subterranean plant parts, describes three new species which occur along with *R. solani*. From the work of Briton-Jones (1924), however, it would appear that the differences used by Matz in separating species are not greater than those between several cultures of the same strain of *R. solani* grown under different conditions. Most of the comparative studies on Rhizoctonia have been made with strains isolated from angiosperms.

Inoculation tests on coniferous seedlings with a number of miscellaneous fungi are reported in the papers of Hartley, Merrill, and Rhoads (1918) and Hartley (1921). More recently, Rathbun-Gravatt (1925) has brought together the results of direct stem-inoculation tests with more than one hundred different strains of fungi. Of these the author finds a few species of Fusarium and certain strains of *Corticium vagum*, *Pythium debaryanum*, *Pythium aphanidermatum* (Eds.) Fitz., and *Botrytis cinerea* Pers., capable of causing damping-off. The last three organisms, together with *Fusarium sporotrichioides* Sherb., are the most virulent. Further tests with this method are reported by Eliason (1928).

In summary, then, *Phytophthora fagi* and *Fusarium* spp. are reported as the chief causal organisms in Europe, and *Pythium debaryanum*, *Corticium vagum*, and *Fusarium* spp. in the United States. Hartley (1921) presents considerable evidence in support of his conclusion that these last-mentioned fungi are equally important as the chief causal organisms involved in the damping-off diseases of conifers in this country.

Many investigators have concerned themselves with attempts to control damping-off of conifers. While various methods have been employed, the most satisfactory results have been obtained through chemical treat-

ment of the soil. A comprehensive discussion of this phase of control may be found in the paper by Hartley and Pierce (1917), who bring together not only the results of their own extensive experiments but also the published results of other investigators. Only the more important papers dealing with the use of sulfuric acid, formaldehyde, zinc chloride, copper sulfate, aluminum sulfate, and various mercury compounds, are discussed here.

Spaulding (1908, 1914), in experiments begun in 1905, devised the sulfuric-acid treatment, while Hartley (1912 b) made the first extensive tests with this material. Other publications by Hartley (1912 a, 1915), Hartley and Merrill (1914), and Hartley and Pierce (1917), have shown that of approximately fifty materials tested, sulfuric acid gave the most promising results. Not only was the acid effective in controlling the disease, but it reduced the number of weeds and caused increased growth of the seedlings. However, it was ineffective in alkaline soils, and in some nurseries caused chemical injury which necessitated frequent watering of the beds during the period of germination and emergence. In a more recent consideration of the original data, Hartley (1921) concludes that the necessity for this frequent watering can be eliminated if in each locality the minimum quantity of acid which will give reasonable control is employed. Later reports on the use of sulfuric acid are those of Hansen and others (1923), Toumey and Li (1924), Delevoy (1926), Wiant (1927), and May and Young (1927).

Early experiments with formaldehyde were made by Spaulding (1908, 1914), Jones (1908), and Gifford (1911). Hartley and his colleagues (Hartley, 1912 a; Hartley and Merrill, 1914; Hartley and Pierce, 1917) found formaldehyde effective, but less satisfactory than sulfuric acid. Similar reports are made by Hansen and others (1923) and by Toumey and Li (1924). The latter authors, as well as previous workers, noted considerable reduction in emergence on the formaldehyde-treated beds. May and Young (1927) reported that formaldehyde was superior to sulfuric acid. From their published data it appears that under the same conditions no treatment was made on more than a single plat.

Zinc chloride and copper sulfate were found effective by Hartley and his associates (Hartley, 1912 a; Hartley and Merrill, 1914; Hartley and Pierce, 1917), and were recommended by them for use on alkaline soils where sulfuric acid could not be employed. Hansen and others (1923) also report tests with these two materials, but unfortunately these investigators introduced a large number of varying factors into their experiments without providing for numerous replications.

Aluminum sulfate was successfully employed for the control of damping-off of conifers under the direction of Hartley (Anonymous, 1926). Similar results with this material were obtained by the writer (Wiant, 1927).

Hartley and Merrill (1914) report unfavorable results from a limited

number of tests with mercuric chloride. In a later paper, Hartley (1915) points out that this material caused severe chemical injury, as was evidenced by the killing of dormant seed. The writer (Wiant, 1927), however, using much weaker concentrations than those employed by Hartley, obtained excellent results with mercuric chloride on coniferous seedbeds.

During the past several years, a number of commercial fungicidal preparations have appeared which contain as their toxic principle various organic mercury compounds, such as chlorophenol mercury, cyancresol mercury, and nitrophenol mercury. The first published account of the use of these preparations for the control of damping-off of conifers is that of the writer (Wiant, 1927), who points out their effectiveness when used both in solution and in the dry form. May and Young (1927), whose paper has already been referred to, used several of the organic mercury preparations in small-scale experiments.

The following papers deal with the use of a number of organic and inorganic mercury compounds for controlling Rhizoctonia causing damping-off of vegetable seedlings and brown patch of turf. Rosen (1924) states that chlorophenol mercury is effective in controlling Rhizoctonia damping-off of cotton. Gloyer and Glasgow (1924, a and b) found that mercuric chloride successfully controlled Rhizoctonia attacking cabbage. Clayton (1926), using chlorophenol mercury, cyancresol mercury, mercuric chloride, and nitrophenol mercury, failed to control damping-off in cruciferous seedbeds, but suggests as a possible explanation the fact that the first applications were made two weeks after emergence had occurred. H. E. Thomas (1927) found chlorophenol mercury and mercuric chloride effective for controlling Rhizoctonia damping-off in flats of cabbage and tomato. Small (1927) reports success with chlorophenol mercury in controlling foot-rot of tomato caused by Rhizoctonia. Coons and Stewart (1927) found that dusting sugar-beet seed-balls with chlorophenol mercury reduced the infection from soil-borne Rhizoctonia. A similar effect from liquid treatments with the same chemical on garden beets is reported by McWhorter (1927).

Both mercurous chloride and mercuric chloride were unsuccessfully employed in preliminary experiments by Piper and Oakley (1921) and by Oakley (1924) for controlling the brown patch of turf. Godfrey (1925) later demonstrated the effectiveness of chlorophenol mercury against that disease. Monteith (1925 a) reports similar results, but finds nitrophenol mercury ineffective. Monteith (1925 b, 1926), and Monteith and Harmon (1927), report extensive tests with a large number of organic and inorganic compounds of mercury, many of which, including mercuric chloride, were effective. They conclude, however, that mercurous chloride was the most satisfactory from the standpoints of control, toxicity, and cost.

PRESENT INVESTIGATIONS

DESCRIPTION OF THE NURSERIES

The Ithaca nursery consisted of twelve seedbeds located on the experimental farm of Cornell University. The beds were 4 by 12 feet in size, and were completely inclosed by a framework of wood which was covered with wire mesh. An additional half-shade frame was placed over the tops. The nursery was bordered by buildings and trees on three sides, but was open toward the south. The soil is a silty loam.

The Keene nursery, where most of the present investigations were conducted, is a large commercial nursery located near the city of Keene in southern New Hampshire. Situated along the southeastern terrace of the Ashuelot River Valley, this nursery is some thirty feet above the level of the valley bottom and is consequently well exposed to the prevailing west winds. Thus, excellent air drainage is provided over the greater part of the seedbed area. The soil is a medium sandy loam containing less than 3 per cent of organic matter and with a reaction of pH 4.8 to 5.2. This loam is underlain by nearly pure sand to a depth of eighteen inches, where it grades off into a coarse gravel. At about eight feet below the surface there is a layer of blue clay. Soil drainage is therefore rapid.

There are sown annually at this nursery approximately 50,000 square feet of seedbed, 70 per cent of which is devoted to norway spruce (*Picea Abies* [L.] Karst.), white pine (*Pinus strobus* L.), and red pine (*Pinus resinosa* Ait.). The remainder is used for colorado spruce (*Picea pungens* Engelm.), white spruce (*Picea glauca* Voss.), scotch pine (*Pinus sylvestris* L.), austrian pine (*Pinus nigra* Arnold var. *austriaca* Aschers. & Graebn.), and a number of other species, including balsam fir (*Abies balsamea* [L.] Mill.), concolor fir (*Abies concolor* Lindl. and Gord.), european larch (*Larix decidua* Mill.), engelmann spruce (*Picea Engelmanni* Engelm.), mugho pine (*Pinus mugo* Turra var. *mughus* Zenari), douglas spruce (*Pseudotsuga taxifolia* Britt.), american arborvitae (*Thuja occidentalis* L.), oriental arborvitae (*Thuja orientalis* L.), common hemlock (*Tsuga canadensis* [L.] Carr.), and carolina hemlock (*Tsuga caroliniana* Engelm.). From several points of view, norway spruce is the most important species grown.

The method of preparing the seedbeds is as follows. After the land has been plowed, harrowed, and fertilized, it is raked over by hand. Beds 4 feet wide and from 50 to 300 feet long are then laid out and separated from one another by 18-inch paths. The beds are raked, rolled, and marked out into drills running crosswise of the beds, 4 inches apart. Except for a few beds which are purposely arched in the middle, the level of the beds is little, if any, higher than that of the paths. Seeding is accomplished by walking down through the middle of the bed and sifting the seed into the drills from a long-necked bottle. A second rolling tends to close the drills and leave the seed covered with a thin layer of soil. Wherever this fails,

a little soil from the paths is sprinkled on. The beds are then covered with a single thickness of burlap cloth, which is removed when emergence begins. The burlap is held in place and the beds are protected by wooden lath-covered frames placed flat upon the beds. These are provided with hinged legs, which can be used to elevate the frames some 8 or 10 inches above the surface of the beds after the burlap has been removed and the seedlings have begun to appear. The strips of lath covering the frames are spaced 1 inch apart, so that partial shade is provided during the two summers that the seedlings are left in the beds.

PREVALENCE OF THE DISEASE

The seedbeds at the Ithaca nursery were used for one season only. In order to insure damping-off, the beds were artificially infested with a culture of Rhizoctonia known to be pathogenic to red pine, the one species grown here. The disease was therefore extremely severe at this nursery. An almost complete loss of the seedlings resulted on the untreated plats.

From the time when the Keene nursery was established in 1906, until the spring of 1924, no serious damping-off was noted there by those in charge. In 1924 the disease appeared rather generally but caused heavy losses only in one corner of the nursery, where from 15 to 20 per cent of damping-off occurred in beds of white pine and norway spruce.

In 1925 the disease was very destructive in the red-pine and norway-spruce beds, resulting in losses of approximately 50 per cent and 35 per cent, respectively. These figures are rough estimates arrived at by the writer from observations on the final stand as noted during the summer of 1926. A section of the red-pine beds as they appeared when the above estimates were made is illustrated in Plate I, 1. The loss in the area photographed was practically 70 per cent. These particular beds were located near the area in which damping-off was severe in 1924.

The major part of the spring sowing of 1926 was made on a section of the nursery which had never before been used for seedbeds. Contrary to expectations, damping-off was far more severe than at any previous time in the history of the nursery. The seedlings on many beds were almost completely destroyed. Such an area of norway spruce is illustrated in Plate I, 2. The disease became evident with the initial appearance of seedlings of the earliest sowing, and continued to occur throughout the general period of emergence and for several weeks thereafter. It was manifested by an extreme reduction in emergence (particularly in norway spruce), and by the usual damping-off of seedlings after they had appeared above ground. In the case of white pine many seedlings were evidently destroyed by a rotting of the young roots when the stem tissues were already beyond the succulent stage.

The regular 1927 seeding was made for the most part during the preceding autumn, in a section of the nursery several hundred yards distant from the

area where the disease had been prevalent in previous years. The incidence of disease was low in contrast with that of 1925 and 1926. The beds used for the experiments on control were, on the other hand, spring-sown and purposely located in infested areas. Severe damping-off occurred in most of the check plats. The relative importance of the two factors, location and time of seeding, is considered elsewhere in this work, but it is interesting to note here the pronounced difference in prevalence of the disease between the two areas just referred to. The losses from damping-off at the Keene nursery for the three years 1925 to 1927 inclusive, are summarized by species in table 1, those for the experimental beds where soil treatments were employed not being included.

TABLE 1. LOSSES FROM DAMPING-OFF AT THE KEENE NURSERY FROM 1925 TO 1927 INCLUSIVE

Species sown	Year					
	1925		1926		1927	
	Area sown, expressed in number of 4-by-12-foot beds	Estimated percentage of damping-off after emergence	Area sown, expressed in number of 4-by-12-foot beds	Estimated percentage of damping-off after emergence	Area sown, expressed in number of 4-by-12-foot beds	Estimated percentage of damping-off after emergence
Austrian pine	10	.....	13	None	28	5
Mugho pine	19	5–10	14	Severe*	5	Trace
Red pine	119	50	88	10–15	142	7
Scotch pine	25	5	42	None	32	5
White pine	294	3–5	445	30	192	5
Colorado spruce	6	80	38	45	45	10
Engelmann spruce	.....	.....	10	None	4	Trace
Norway spruce	225	35	311	50	456	7
White spruce	18	.....	33	Severe*	26	Trace
Douglas spruce	168	.....	.....	.....	.....	.....
Balsam fir	30	.....	3	None	.....	.....
Concolor fir	.....	.....	12	None	.....	.....
European larch	.....	.....	42	Severe*	18	None
American arborvitae	20	.....	31	None	13	None
Oriental arborvitae	17	.....	17	Trace	13	None
Common hemlock / Carolina hemlock	27	.....	29	None	46	None

* The seed was of low viability, and so counts were not made.

The data for 1925 were estimated from the final stand one year after seeding, and are therefore only approximations. The figures for 1926 and 1927 were based partly upon estimates from careful observations during

the season, and partly upon counts. They represent percentages of damping-off after emergence, on the basis of plants emerged, and are therefore low.

It is apparent that the disease has been most prevalent on norway spruce, white pine, and red pine — the three major species here grown — and on colorado spruce. Mugho pine, white spruce, and european larch suffered serious losses in 1926. The disease has been found also on scotch and austrian pine, as well as a trace on oriental arborvitae. Much of the variation between the same species in different years, and between different species in the same year, can be accounted for by the location of the seedbeds.

From these data one can obtain some idea of the losses that may result from this disease. There is in the first place a reduction in the quantity of salable stock. The tendency of most nurserymen in this situation is to sow large numbers of surplus seedbeds in order to maintain the production. This involves the added cost of seed, the price of which varies from $1 to $3 per pound for norway spruce and white pine, to $15 per pound for red pine; the increased cost of the seeding operations; and, most important of all, the greater expense of weeding and caring for the seedbeds during the two years between sowing and transplanting. There must then be considered the irregularity of production, which becomes particularly important in those cases in which long-term contracts are made for annually supplying large quantities of nursery stock. The combination of these factors serves to make such epiphytotics a problem of major importance.

ETIOLOGY

Isolation of organisms from diseased seedlings

Wherever possible, seedlings were collected for purposes of culturing the pathogenes soon after the seedlings exhibited symptoms of the disease. When the field work was first begun, the method of handling these was as follows. Short pieces of the stem were removed in such a manner as to include a part of the healthy tissue and an adjacent bit of the diseased tissue. These were immersed in a 1–1000 solution of mercuric chloride for one minute, rinsed in distilled water, and transferred to petri dishes of potato-dextrose agar. After several days, transfers were made from the margin of the advancing mycelium to other dishes of the same medium. The organisms were then transferred to agar slants. Since Alternaria, Fusarium, Pythium, Rhizoctonia, and other common fungi grew well on this agar, it was adopted as the standard medium throughout the course of these studies. Common saprophytes, such as species of Penicillium and Mucor, were discarded as soon as they were detected.

The method just described was somewhat modified in later work at the Keene nursery, the chief variation consisting in washing the sections of diseased stems with sterile water from a wash bottle and plating them

Plate I

DAMPING-OFF LOSSES AT THE KEENE NURSERY

1, Section of 1925 seeding of red pine. All seedlings were destroyed in the immediate foreground and in scattered spots throughout the beds. Losses were estimated at 70 per cent. (Photographed at end of second growing season)

2, Section of 1926 seeding of norway spruce. A reduction in stand of approximately three-fourths of a million seedlings occurred on this area. (Photographed four months after seeding)

1

2

directly. The greater rapidity of this method permitted more extensive examinations than could otherwise have been made. The method was frequently checked by similarly examining sections of healthy seedlings. Such organisms as did occur on these plates were common saprophytes. Direct microscopic examination of diseased tissues made at frequent intervals also served as a check on the method of isolation here employed.

The results obtained from the Ithaca nursery are summarized in table 2 on the basis of fungi isolated from both untreated beds and those treated with chemicals for controlling the disease. The seedlings, all of which were red pine, were collected during the last three weeks of June, 1925.

TABLE 2. SUMMARY OF ISOLATIONS FROM DISEASED SEEDLINGS AT THE ITHACA NURSERY

Treatments		Number of seedlings plated	Fungi isolated					
Materials used	Number of applications		Rhizoctonia	Pythium	Fusarium	Alternaria	Phoma	Unidentified
Untreated	.....	75	63	7	2	0	0	3
Sulfuric acid	1	56	31	8	3	8	3	3
Uspulun	2	31	0	20	1	5	3	2
Uspulun	3	33	0	12	1	5	5	10
Uspulun	6	29	1	10	2	7	4	5
Mercuric chloride	5	47	1	25	2	6	2	11
Bayer compound	5	48	8	23	2	4	1	10
Germisan	2	27	17	4	0	0	0	6
Total	.....	346	121	109	13	35	18	50

The data presented in table 2 indicate that Rhizoctonia and Pythium were the two fungi most frequently isolated at the Ithaca nursery, while Alternaria, Phoma, and Fusarium also were found associated with diseased seedlings. It is interesting to note that the seedlings from the untreated beds, and those treated with sulfuric acid and Germisan, respectively, yielded a much higher percentage of Rhizoctonia than of Pythium, while the reverse is true for those beds treated with other materials. Since the beds were artificially infested with Rhizoctonia, it would seem likely that under natural conditions Pythium would have been the most important organism involved in these beds.

During the season of 1926, extensive isolations were made at the Keene nursery from seedlings representing a large number of infection foci in beds of austrian, mugho, red, and white pine, colorado and norway spruce, and oriental arborvitae. Collections were made from June 1 to August 2, the majority, however, being made during the month of June. It is

believed that the seedlings selected were typical in all respects of the large numbers which were destroyed during the epiphytotic of that season.

During the following season, collections were made from June 4 to June 23. As noted in table 1, the average loss from damping-off was low in the regular nursery seedbeds of that year, so that while many seedlings were collected from those beds the majority were taken from the experimental beds where the incidence of the disease was very high.

In addition to the seedlings collected at Keene during the season of 1927, specimens were at the same time received from a number of other nurseries as follows: white and red pine from Gerrish, New Hampshire; red pine from Milton, Pennsylvania; austrian and scotch pine from Amherst, Massachusetts; austrian pine, and colorado, engelmann, and norway spruce, from Framingham, Massachusetts; norway spruce from Essex Junction, Vermont; and japanese larch, norway spruce, and red, white, and scotch pine, from St. Williams, Ontario. These were collected from June 11 to June 21, and were for the most part in good condition when received. Isolations were made within a few days from the time when the seedlings were collected. While comparatively small numbers of the seedlings were examined in each case, the results give some indication of the organisms associated with the disease at these nurseries. A summary of these results, together with those from the Keene nursery in 1926 and 1927, is given in table 3.

TABLE 3. SUMMARY OF ISOLATIONS FROM THE KEENE AND OTHER NURSERIES

Location of nursery	Number of diseased seedlings	Number showing no growth	Number of fungi isolated						
			Rhizoctonia	Rhizoctonia and Fusarium	Rhizoctonia and Alternaria	Fusarium	Alternaria	Pythium	Penicillium or Mucor
Keene, N. H. (1926)	584	61	320	.....	.....	.....	.....	.....	101
Keene, N. H. (1927)	734	30	594	.....	.....	6	.....	.....	79
Milton, Pa.	45	.....	16	17	9	1	1	.....	1
Gerrish, N. H.	105	.....	77	18	.....	2	.....	.....	7
Amherst, Mass.	104	2	45	28	.....	14	.....	.....	15
Framingham, Mass.	85	.....	3	.....	.....	51	.....	.....	.....
Essex Junction, Vt.	75	.....	1	.....	.....	37	.....	.....	.....
St. Williams, Ont.	93	.....	Few	.....	.....	Very many	.....	Many	.....

The results from the Keene nursery are very striking, in that Rhizoctonia was isolated from the vast majority of more than a thousand seedlings. Although particular care was used in searching for Pythium, Fusarium, Alternaria, and other fungi which were found associated with the diseased seedlings at the Ithaca nursery, these organisms were rarely found at the Keene nursery in seedlings which were examined soon after they exhibited the first symptoms of the disease. Direct microscopic examination of such plants showed the tissues to be filled with the characteristic mycelium of Rhizoctonia. A number of the fungi noted above were

obtained in a few cases from white-pine seedlings that had been killed several weeks previous to the time of collection. These miscellaneous organisms undoubtedly represented saprophytic development in seedlings already killed by the damping-off fungus.

The data in table 3 further indicate that Rhizoctonia was the chief organism associated with diseased seedlings obtained from the Milton, the Gerrish, and the Amherst nurseries, although Fusarium and Alternaria were both found to a lesser extent in one or more cases. Microscopic examination, however, showed that Rhizoctonia was more abundant than the other two organisms in the diseased tissues. The data for the other three nurseries are less detailed, but indicate that Fusarium was predominant in the diseased seedlings. Pythium was abundant in the seedlings from the St. Williams nursery. These specimens were in a rather advanced state of decay when they were received.

Inoculation and reisolation studies

When inoculation studies were first begun during the winter of 1925–26, the method employed was that of growing the seedlings in three-inch pots of unsterilized soil to which small amounts of inoculum had been added. Five pots were used for each organism, and three served as checks. The inoculum was prepared by growing the fungus for three or more weeks in flasks of steamed wheat kernels. Ordinary tap water was employed in sprinkling. In these experiments, considerable damping-off occurred in many of the check pots. Because of this, all subsequent tests were made in steam-sterilized soil and the number of check pots was increased from three to five. The pots were filled two-thirds full with a rather heavy loam, and steam-sterilized for two to three hours at 25 pounds pressure, following which 5 grams of inoculum was added to each pot. The inoculum was covered with a half-inch layer of a sterilized muck and sand mixture, and sufficient seed was sown to produce from twenty to forty seedlings per pot. The seed was covered with a thin layer of either sand or muck. During the winter of 1926–27, only distilled water was used in sprinkling, while ordinary tap water was employed during the following season.

For the first two winters the mean daily temperature of the greenhouse varied from 65° to 75° F., while for the third season automatic controls maintained a fairly uniform temperature of approximately 65° F.

Since the methods thus varied somewhat during the three winters, it should be pointed out that all of the Pythium strains and most of the other fungi from the Ithaca nursery were tested during 1925–26. The majority of the Rhizoctonia strains from the Keene nursery were tested in 1926–27, and all other fungi in 1927–28.

The results obtained from inoculation experiments with more than a hundred strains of fungi are brought together in table 4. While records

TABLE 4. RESULTS OF INOCULATION EXPERIMENTS WITH STRAINS OF FUNGI ISOLATED FROM DISEASED CONIFEROUS SEEDLINGS

Fungus tested	Conifer from which strain was isolated and on which it was tested*	Source†	Total number of strains tested	Number of strains showing differences in final stand as indicated below, between inoculated and check pots (differences expressed in percentage of seed sown)‡								
				80 to 90 per cent	70 to 80 per cent	60 to 70 per cent	50 to 60 per cent	40 to 50 per cent	30 to 40 per cent	20 to 30 per cent	10 to 20 per cent	Not significant
Rhizoctonia	R.P.	K	33	2	2	5	4	6	2	2	.....	10
Rhizoctonia	W.P.	K	20	.....	.....	.....	3	6	8	.....	.....	3
Rhizoctonia	M.P.	K	5	.....	.....	.....	.....	.....	.....	1	2	2
Rhizoctonia	N.S.	K	4	.....	.....	.....	.....	1	2	1	.....	.....
Rhizoctonia	A.P.	K	3	.....	.....	.....	1	1	1	.....	.....	.....
Rhizoctonia	C.S.	K	4	.....	.....	.....	1	2	1	.....	.....	.....
Rhizoctonia	O.A.V.	K	2	.....	.....	.....	1	.....	1	.....	.....	.....
Rhizoctonia	R.P.	I	9	3	1	.....	1	1	.....	1	.....	2
Rhizoctonia	R.P.	M	3	.....	.....	.....	2	.....	.....	.....	.....	1
Rhizoctonia	R.P.	C	1	.....	.....	.....	.....	.....	.....	1	.....	.....
Rhizoctonia	A.P.	A	2	.....	1	1	.....	.....	.....	.....	.....	.....
Rhizoctonia	A.P.	F	1	.....	.....	1	.....	.....	.....	.....	.....	.....
Rhizoctonia (summary)	.....	.....	87	5	4	7	13	17	15	6	2	18
Pythium	R.P.	I	21	2	5	3	1	.....	1	5	.....	4
Fusarium	R.P.	I	5	.....	.....	.....	.....	.....	.....	.....	.....	5
Alternaria	R.P.	I	3	.....	.....	.....	.....	.....	.....	.....	.....	3
Sterile black mycelium	R.P.	I	3	.....	.....	.....	.....	.....	.....	.....	.....	3

* Abbreviations used are as follows: A.P., austrian pine; C.S., colorado spruce; M.P., mugho pine; N.S., norway spruce; O.A.V., oriental arborvitae; R.P., red pine; W.P., white pine.

† Reference to nurseries are as follows: A, Amherst, Mass.; C, Concord, N. H.; F, Framingham, Mass.; I, Ithaca, N. Y.; K, Keene, N. H.; M, Milton, Pa.

‡ In the case of white pine, based on total viable seed.

were kept of the total emergence, damping-off, and final stand, the last only is included in the table. The difference between the average final stand in the five inoculated and the three or five check pots was thus calculated for each strain and determined in percentage of the total seed sown per pot. Since the viability of the white-pine seed had previously been determined as approximately 30 per cent, the results with this species are based upon the total viable seed.

Where the differences between check and inoculated pots were less than four times the probable error, the results were not considered significant. Such differences were for the most part very small. Those strains which produced a significant difference are grouped into eight classes according to the size of the difference. The figures in each column of table 4 therefore represent the number of strains that produced differences falling within the range of the class values indicated at the head of the column.

The data indicate that the vast majority of the strains of Pythium and Rhizoctonia were highly virulent under the conditions of these tests. This was manifested in the experiments both by reduced emergence and by increased damping-off in the inoculated pots. In the few tests with Alternaria, Fusarium, and an unidentified sterile fungus, these organisms were not found to be pathogenic. Other tests not included in the table indicated that three strains of Alternaria from red pine were non-pathogenic to colorado spruce, austrian pine, and scotch pine, while eight additional strains of Fusarium obtained from scotch and austrian pine were likewise not pathogenic. Of the eighty-seven strains of Rhizoctonia tested, forty-five gave differences of from 30 to 60 per cent while sixteen showed even greater differences in final stand between the inoculated and the check pots. It is interesting to note the somewhat wider range of virulence indicated for the strains on red pine than for those on other species. Similarly, in the tests with Pythium on red pine a wide range of virulence is to be noted.

Reisolations from several hundred diseased seedlings were made throughout the course of these experiments. In most cases the fungus used in the inoculation test was recovered from such seedlings.

The mass of evidence derived from isolation, inoculation, and reisolation studies demonstrates conclusively that Rhizoctonia is the principal pathogene involved in the nurseries where the experiments recorded in this paper were made, and throws additional light upon the question of the etiology of coniferous damping-off in this region.

Comparative study of some strains of Rhizoctonia

As pointed out earlier in this paper, most of the investigations dealing with comparative studies of Rhizoctonia involve strains isolated from angiosperms. Where conifer strains have been employed, attention has been chiefly devoted to inoculation tests performed for the most part on

conifers. Comparatively little study has been made of their morphological characters. In view of the apparent differences between strains isolated from conifers during the course of these investigations, it has seemed desirable to make a comparative study of a number of these strains and a further comparison with several strains isolated from vegetables. The studies were divided into two parts: (1) cross-inoculation tests with thirteen strains on a variety of coniferous and vegetable seedlings; and (2) cultural studies with twenty-three strains on several artificial media. The conifer strains studied were selected as representative of the various types isolated. The several vegetable strains do not represent a wide selection from any particular group of suscepts. The twenty-three strains are listed below, together with the history of each. Those that are starred were used in both cross-inoculation tests and cultural studies, the others in cultural studies only.

Strain no.	Original suscept	Location of diseased plant	Year isolated
141	Austrian pine	Keene, N. H	1926
155	Austrian pine	Amherst, Mass	1927
*160	Austrian pine	Framingham, Mass	1927
*167	Colorado spruce	Keene, N. H	1927
175	Colorado spruce	Keene, N. H	1927
* 77	Norway spruce	Keene, N. H	1926
99	Oriental arborvitae	Keene, N. H	1926
* 13	Red pine	Ithaca, N. Y	1925
84	Red pine	Keene, N. H	1926
177	Red pine	Keene, N. H	1927
172	Red pine	Milton, Pa	1927
190	Red pine	Milton, Pa	1927
191	Red pine	Milton, Pa	1927
*192	Red pine	Milton, Pa	1927
*156	Scotch pine	Amherst, Mass	1927
158	White pine	Concord, N. H	1927
*187	White pine	St. Williams, Ont	1927
* 1	Cabbage	Eden, N. Y	1920
*184	Lobelia	Ithaca, N. Y	1927
*183	Pepper	Schenectady County, N. Y	1926
*193	Potato	Ithaca, N. Y	1927
*195	Sugar beet	Rocky Ford, Colo	1927
*196	Sugar beet	Rocky Ford, Colo	1927

The potato strain was isolated from sclerotia found on the surface of tubers; the lobelia strain from lesions on the stems and roots of that plant; the beet strains from lesions on the fleshy roots, those caused by strain 196 being of the dry-rot-canker type described by Richards (1921). All other strains were obtained from diseased seedlings.

With the exception of the following, all strains were isolated by the writer: the cabbage strain, which was a subculture of one isolated by

Gratz (1925); the lobelia strain, isolated by S. C. Teng; the beet strains, isolated by Dewey Stewart; and the pepper strain, isolated by Dr. H. E. Thomas.

Cross-inoculation tests

Thirteen strains of Rhizoctonia, including seven isolated from conifers and six from vegetables, were used in cross-inoculation studies. The species on which they were tested represent a selection from the more important species of plants commonly attacked in the seedling stage by Rhizoctonia. With two exceptions, white pine and lobelia, they include all of the suscepts from which the strains were originally isolated.

The methods employed were similar to those already described for the 1927–28 inoculation tests on conifers. The temperature of the greenhouse was maintained at approximately 65° F. throughout the course of the experiment. In testing each species of vegetable, three five-inch pots were inoculated with a given strain of the fungus, while an equal number were left as checks. The tests on conifers were conducted as previously described. The beet seed-balls were pasteurized at 60° C. for ten minutes on two successive days, in order to avoid infection from seed-borne fungi. It was not possible to carry out all of the tests simultaneously, but special efforts were made to maintain fairly uniform conditions throughout the course of the experiment.

The results of the cross-inoculation tests are found in tables 5 and 6, where the average difference between check and inoculated pots is given for each strain, this difference being expressed in percentage of seed sown. For sugar beets, however, it was found that in the check pots approximately one-third more seedlings appeared than the number of seed balls planted. Differences in final stand are therefore based on this expected stand, since it is impossible to know the exact number of seeds contained in a given number of seed balls. Because of the rather limited number of pots used in each test, differences that are less than four times the probable error are not considered significant.

As shown by the data in table 5, four of the seven strains from conifers (160, 77, 156, and 13) were highly virulent on conifers. In one case only (strain 77 on austrian pine) the difference is not significant — due, however, to contamination in several of the check pots. Strain 192 did not appear to be pathogenic to conifers. Strains 187 and 167, particularly the latter, were of relatively low virulence, as shown by the low average for all species tested and by the fact that in two and three cases, respectively, the differences are not significant.

Of the six vegetable strains tested on conifers, strains 183 and 196 were highly virulent, strains 1 and 184 were mildly virulent, strain 195 was of very low virulence, and strain 193 was non-pathogenic.

There is little evidence from the data in table 5 to indicate that a given strain is more virulent than others on the species from which it was origi-

TABLE 5. RESULTS OF INOCULATION TESTS ON SPECIES OF CONIFER SEEDLINGS WITH THIRTEEN STRAINS OF RHIZOCTONIA

Strain no.	Plant from which strain was isolated	Average difference between check and inoculated pots for each strain, expressed in percentage of seed sown					
		On austrian pine	On red pine	On colorado spruce	On norway spruce	On scotch pine	Average of the five
160	Austrian pine	79.2 ± 3.3	61.2 ± 3.9	65.6 ± 9.5	50.0 ± 2.4	36.0 ± 4.7	58.4
77	Norway spruce	54.4 ± 15.1	54.8 ± 7.9	62.4 ± 3.9	47.2 ± 2.6	43.2 ± 2.1	52.4
156	Scotch pine	77.6 ± 4.2	30.4 ± 3.5	56.4 ± 8.4	32.4 ± 5.1	53.2 ± 1.8	50.0
13	Red pine	68.8 ± 6.3	52.8 ± 4.7	40.0 ± 4.6	38.4 ± 1.5	36.8 ± 3.8	47.4
187	White pine	62.4 ± 11.7	−1.2 ± 6.1	29.2 ± 8.6	23.6 ± 3.8	42.0 ± 2.5	31.2
167	Colorado spruce	47.2 ± 4.2	19.2 ± 5.6	26.0 ± 7.2	28.0 ± 1.6	15.2 ± 5.5	27.1
192	Red pine	−1.6 ± 2.5	8.4 ± 4.3	−19.6 ± 11.2	4.0 ± 4.0	6.8 ± 5.1	−0.4
183	Pepper	81.6 ± 2.5	38.4 ± 3.8	54.0 ± 10.9	39.0 ± 2.7	32.0 ± 5.0	49.0
196	Sugar beet	60.8 ± 5.6	25.6 ± 5.6	72.4 ± 12.3	41.2 ± 3.4	39.2 ± 4.0	47.8
184	Lobelia	58.4 ± 2.8	20.0 ± 6.0	28.8 ± 7.4	26.4 ± 3.9	21.2 ± 4.4	31.0
1	Cabbage	41.6 ± 4.7	17.2 ± 3.0	23.6 ± 13.1	7.6 ± 5.1	.....	22.5
195	Sugar beet	56.8 ± 6.3	20.0 ± 2.5	11.2 ± 14.1	−4.0 ± 6.5	22.0 ± 6.0	21.2
193	Potato	3.2 ± 2.0	−3.2 ± 7.5	4.4 ± 6.4	3.6 ± 3.7	6.0 ± 4.2	2.8

TABLE 6. RESULTS OF INOCULATION TESTS ON SPECIES OF VEGETABLE SEEDLINGS WITH THIRTEEN STRAINS OF RHIZOCTONIA

Strain no.	Plant from which strain was isolated	Average difference between check and inoculated pots for each strain, expressed in percentage of seed sown						
		On pea	On cabbage	On sugar beet	On eggplant	On pepper	On tomato	Average of the six
187	White pine	88.4 ± 4.1	80.0 ± 3.7	95.1 ± 6.9	49.3 ± 9.3	4.7 ± 3.6	73.3 ± 2.5	65.1
156	Scotch pine	73.4 ± 2.3	77.3 ± 1.8	.....	16.7 ± 3.5	-26.0 ± 11.4	1.3 ± 2.9	28.5
77	Norway spruce	71.7 ± 9.9	26.0 ± 3.2	45.0 ± 9.3	28.0 ± 5.7	-14.0 ± 6.4	-0.7 ± 2.9	26.0
160	Austrian pine	68.4 ± 1.1	31.3 ± 6.0	9.9 ± 6.0	14.0 ± 5.4	-2.7 ± 1.8	10.0 ± 3.9	21.8
167	Colorado spruce	43.3 ± 9.2	18.7 ± 1.3	45.0 ± 10.4	2.7 ± 3.9	6.0 ± 6.8	8.0 ± 2.5	20.6
13	Red pine	41.7 ± 7.9	17.3 ± 2.2	6.9 ± 9.3	2.6 ± 5.2	5.3 ± 3.9	3.3 ± 4.0	12.9
192	Red pine	30.0 ± 6.4	32.0 ± 4.8	-23.1 ± 9.6	7.3 ± 6.1	1.3 ± 3.1	-1.3 ± 3.1	7.7
196	Sugar beet	98.4 ± 1.1	82.0 ± 3.4	81.0 ± 9.3	64.0 ± 3.1	32.7 ± 8.7	62.7 ± 2.2	70.1
183	Pepper	85.0 ± 3.2	69.3 ± 6.8	75.0 ± 7.0	67.3 ± 5.9	57.3 ± 6.5	45.3 ± 3.1	66.5
1	Cabbage	83.5 ± 3.0	90.0 ± 2.1	66.9 ± 2.4	32.6 ± 10.7	0.0 ± 2.6	5.3 ± 5.1	46.4
195	Sugar beet	8.3 ± 4.2	27.3 ± 3.2	99.0 ± 4.2	7.3 ± 4.6	30.0 ± 3.3	16.0 ± 9.3	31.3
184	Lobelia	56.7 ± 4.7	40.0 ± 3.7	32.1 ± 3.9	12.7 ± 3.4	2.7 ± 2.2	0.7 ± 2.7	24.2
193	Potato	35.0 ± 7.3	-9.3 ± 1.6	3.0 ± 3.6	66.0 ± 3.2	-4.0 ± 5.8	52.7 ± 2.7	23.9

nally isolated. The three most virulent conifer strains, however, gave somewhat greater differences than did the most virulent vegetable strain.

Turning attention to the results of inoculation tests on vegetable seedlings, it is apparent from the data in table 6 that greater variation existed in the susceptibility of the different species than was the case with the several conifer species. Pea, cabbage, and sugar beet were distinctly more susceptible than eggplant, pepper, and tomato.[3]

Of the conifer strains tested on vegetables, strain 187 was the most virulent on all species except pepper, on which none of the conifer strains was pathogenic. Strains 156 and 77 were not pathogenic to either pepper or tomato, while strain 167 did not give significant differences over the check in the case of eggplant, pepper, or tomato. Strains 160, 13, and 192 were pathogenic only to cabbage and pea. On the basis of these facts, together with that of the extreme differences in average results, it would appear that strain 187 was the most virulent on vegetables of any of the conifer strains tested, and that strains 13 and 192 were the least virulent.

Two of the strains from vegetables (196 and 183) were, with one exception, highly virulent on all species of vegetables tested. Strain 1 was highly virulent on cabbage, pea, and sugar beet, but was not pathogenic to eggplant, pepper, and tomato. Strains 195, 184, and 193 were not pathogenic to three suscepts each. The strains from cabbage, sugar beet, and pepper were the most virulent of any of the vegetable strains on the same species of vegetable from which they were respectively isolated. With one exception (strain 187 on sugar beet), the same is true even when all thirteen strains are considered.

From the tests with the thirteen strains on both conifers and vegetables, it would appear that strains 183 and 196 from vegetables were highly virulent on both groups of suscepts. Strain 187 from white pine was highly virulent on vegetables but not of great virulence on conifers. Strains 13, 77, 156, and 160 from conifers were highly virulent on conifers but of low virulence on vegetables. Strains 192 and 167 from conifers, and 193 and 195 from vegetables, were of generally low virulence on both groups of suscepts.

The foregoing relationship is illustrated diagrammatically in figure 1, but, since only the average figures from the last column in tables 5 and 6 are plotted, the diagram should be interpreted in the light of the preceding discussion.

Cultural studies

Among the numerous strains of Rhizoctonia isolated from conifers, it was observed that marked and more or less constant differences were maintained through successive transfers in tubes of potato-dextrose agar. This led to the study of a number of these strains in petri-dish cultures on cornmeal, oatmeal, potato, potato-dextrose, malt-extract, Czapek's, and

[3] Tests were conducted also with potato. Severe infection occurred with strain 183, and slight infection with strain 193. In view of the extreme variation in soil moisture, the writer does not attach particular significance to the negative results obtained with the other strains.

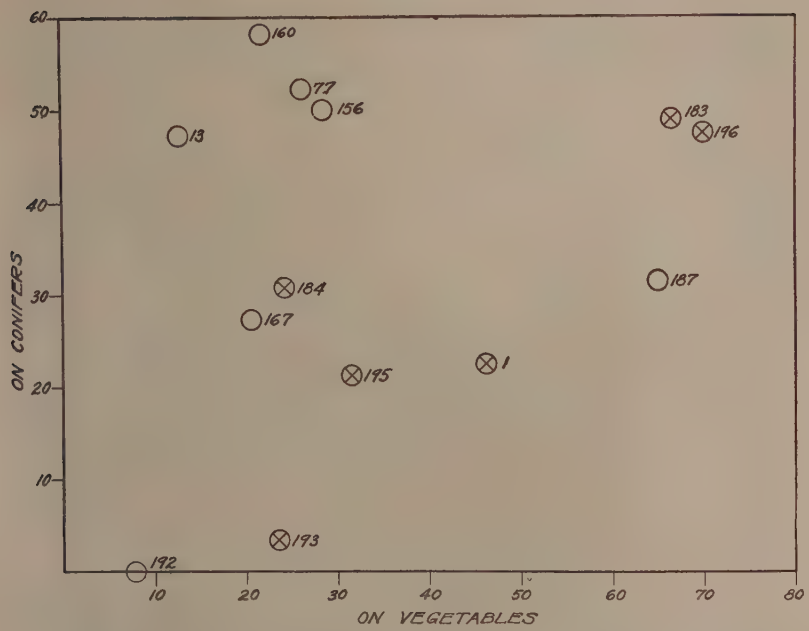

FIGURE 1. CROSS-INOCULATION TESTS WITH THIRTEEN STRAINS OF RHIZOCTONIA

The strains from conifers are designated by a plain circle, those from vegetables by a cross within a circle. The vertical scale represents the average difference between inoculated and check pots for each strain tested on conifers, this difference expressed in percentage of seed sown. The horizontal scale similarly represents the results of the tests on vegetable seedlings. (Data from last column of tables 5 and 6)

beef-peptone agars. Of these media, Czapek's, malt-extract, potato-dextrose, and cornmeal agars were selected as the standard media for later studies.[4] The twenty-three strains of Rhizoctonia already referred to

[4] Formulae of the four media:

Czapek's synthetic-solution agar (Waksman, 1916, p. 113)

Distilled water	1000 cc.
$MgSO_4$	0.50 gm.
K_2HPO_4	1.00 gm.
KCl	0.50 gm.
$FeSO_4$	0.01 gm.
$NaNO_3$	2.00 gm.
Cane sugar	30.00 gm.
Agar	15.00 gm.

Malt-extract agar (Baxter, 1925, p. 528)

Distilled water	1000 cc.
Malt extract	25 gm.
Agar	25 gm.

Potato-dextrose agar

Distilled water	1000 cc.
Pared potatoes	200 gm.
Dextrose	10 gm.
Agar	15 gm.

Potatoes cooked until soft, and broth filtered through cheesecloth

Cornmeal agar

Distilled water	1000 cc.
Cornmeal	30 gm.
Agar	15 gm.

Maintained in water bath at 58° C. for one hour before adding agar

All media were boiled until the agar was dissolved, followed by a thirty-minutes heating in an Arnold sterilizer and slow cooling to permit settling and removal of sediment. Prior to pouring the plates, melting was accomplished by another thirty-minutes exposure to live steam. Determinations made by the electrometric method after the agars had finally solidified, indicated the following pH values for the different media: Czapek's, -6.82; potato-dextrose, -5.75; malt-extract, -4.87; cornmeal, 5.86.

were then grown in triplicate on each of these four media for five weeks at 25° C. Conspicuous differences between strains were evident on Czapek's agar, and to a lesser extent on malt-extract and potato-dextrose agars, while on cornmeal agar so little differentiation occurred that the cultures on that medium were discarded.

The four strains 77, 99, 141, and 177, which were selected as representing the majority of those isolated at the Keene nursery, were indistinguishable from one another on all three media. The principal characters of these strains are: on Czapek's agar, growth poor, sclerotia absent, the aerial hyphae tending to form irregular cottony tufts which are somewhat appressed to the surface of the agar; on malt-extract and potato-dextrose agars, growth abundant, presenting in general a buckthorn brown[5] color on the former and mummy brown on the latter; sclerotia formed on those two agars mostly less than one millimeter in diameter, indefinite in structure, for the most part only denser tufts of the fairly abundant aerial mycelium and profusely scattered over the agar; on malt-extract agar forming a few scattered sclerotia several millimeters in diameter, lighter in color than the mycelium and distinct from the aerial part of the latter (Plate II, 1–3).

A number of other strains (13, 84, 158, 160, 172, 190, 191, and 195) were similar to the above in many characters but differed from them and from one another in certain minor respects.

Several strains very distinct from all others encountered are represented in these studies by strain 156. The outstanding characters of this strain are: formation on Czapek's agar of numerous small compact sclerotia one millimeter or less in diameter and almost black in color, distinct or occasionally fused into oblong masses, always in striking contrast to the white or faintly brown, thin, opaque mycelial membrane; less abundant growth on malt-extract and potato-dextrose agars, with no evidence of a mycelial membrane and with sclerotia fewer and chiefly confined to the periphery of the agar, showing more pronounced fusion into masses on malt-extract agar (Plate II, 4–6).

An outstanding feature occurs in all of the other strains studied, namely, the formation on Czapek's agar of a very dense mycelial membrane on the surface of the medium. These strains vary widely, however, in other respects, as, for example, color, number, size, and type of sclerotia, and abundance of mycelium. Thus, strains 155 and 187 (Plate II, 10–12), which appear identical with the others, are characterized on the three agars by the nearly complete absence of conspicuous sclerotia and by the formation of a light-colored mycelium. Strains 167 and 175, which are believed to represent a considerable number of those isolated from conifers, are somewhat similar to strains 155 and 187 but differ in the formation on malt-extract and potato-dextrose agars of papery sclerotia which are

[5] Colors according to Ridgway (1912).

light in color and are loosely fused with the abundant aerial mycelium (Plate II, 7–9). The four strains 1, 183, 184, and 193, on the contrary, form abundant distinct sclerotia (Plate III, 1–9). Strains 192 and 196 are intermediate in character between these four and the other four noted above (155, 187, 167, and 175) (Plate III, 10–12).

Conclusions from the cross-inoculation and cultural studies

The most outstanding fact brought out by these experiments is that many of the strains of Rhizoctonia isolated from conifers differ strikingly in their virulence and in their morphological characters as expressed by growth on certain artificial media. Apparently the conifer strains differ as much from one another as they do from the vegetable strains. The morphological differences, however, are more striking than the differences in virulence noted, and are apparently not correlated with the latter.

In view of the fact that the cultural studies were of a somewhat limited nature, and since only gross morphological features have been considered in the grouping of the strains, it is not proposed to designate the groups with any particular name. With the possible exception of strain 156, which will be studied in more detail later, the strains employed are all considered to fall within the limits of *Rhizoctonia solani* Kühn as the species is now constituted.

CONTROL

The method of control most extensively tested was that of soil treatment with chemicals. A limited amount of information was obtained also on the effect of adding muck to the soil, and on the relation of fall seeding to the occurrence of the disease.

Application of muck

Because of the low content of organic matter in the soil at the Keene nursery, it has been the practice there to apply large quantities of muck every four or five years to the areas used for seedbeds. General observations by those in charge had suggested the possibility that such applications had been effective in the prevention of damping-off in certain parts of the nursery. Opportunity was afforded to make direct observations on seedbeds of norway spruce sown during the fall of 1926 and the spring of 1927 in a section of the nursery in which muck had never been used. The muck was added in November to several 100-foot beds as a layer several inches thick, after which it was harrowed into the soil. A part of this area was seeded in the fall, and the remainder during the following spring. On the fall-sown beds both with and without muck, approximately 30 per cent of the seedlings that emerged were destroyed by damping-off. More detailed observations were made on the spring-sown beds, where the

following data were obtained from five nine-row plats in each of the treated and untreated areas.

Treatment	Percentage of emergence	Percentage of damping-off after emergence	Final stand
Treated with muck	8.2±0.6	57.2±2.7	148.8±12.9
Untreated	11.2±1.3	55.2±3.6	222.0±34.2

It is apparent that the results obtained with the muck were not significantly different from those where it was not used. Observations covering two years have failed to reveal any convincing evidence that the occurrence of damping-off in different parts of this nursery is associated with applications of muck to the soil.

Fall seeding

The practice of sowing coniferous seedbeds during the fall is followed by many nurserymen. Where practicable it has the advantage of distributing nursery operations more uniformly throughout the season, and thus relieving the congestion of work during the early spring months when transplanting and shipping operations are at their height. To carry out such a program, it is necessary to get the seed into the soil late enough to prevent fall germination and yet early enough to escape the colder weather which would make soil operations impossible. An obvious advantage of the practice is to insure early germination in the spring, and thus provide a longer growing season than would be available for spring-sown stock. Many nurserymen believe that the method has a further advantage in enabling them to avoid the losses from damping-off which are so common on beds sown during the spring, their reason being that by the time weather conditions are most favorable for damping-off the seedlings will have developed far enough to have escaped the disease.

Observations were made for one season, at the Keene nursery, on the relative abundance of the disease in beds sown during the fall of 1926 and in those sown at varying periods during the following spring. The several locations of the seedbeds were designated by letters. Locations B and C were in that part of the nursery where damping-off had been most severe in previous years. They were about 100 yards apart, and each in turn was several hundred yards removed from location A, where most of the beds were sown and where damping-off had never been a serious problem. The part of the nursery designated as location D was intermediate in position between B and C. In table 7 are given the percentages of damping-off that were recorded from the various beds.

TABLE 7. APPROXIMATE PERCENTAGE OF DAMPING-OFF AFTER EMERGENCE IN RELATION TO TIME OF SOWING AT THE KEENE NURSERY DURING FALL AND SPRING, 1926–27

Species sown	Location	November 4–6, 1926		April 1, 1927		April 12, 1927		April 21, 1927		May 6–7, 1927		May 12–16, 1927	
		Number of beds	Per cent	Number of beds	Per cent	Number of beds	Per cent	Number of beds	Per cent	Number of beds	Per cent	Number of beds	Per cent
Colorado spruce	A	20	16	17	5	...	...	8	15	...	...	...	....
Norway spruce	A	232	7	52	5	72	5	84	5	...	...	...	....
Norway spruce	B	20	30	...	...	...	...	...	...	60	58*	...	....
Norway spruce	D	20	20	...	...	...	...	...	...	...	...	...	....
Red pine	A	82	3	17	5	...	...	34	14	...	...	...	....
Red pine	D	9	15	...	...	...	...	...	...	...	...	...	....
Red pine	C	...	...	...	...	...	...	...	...	...	...	83	61*
White pine	A	109	5	...	...	...	...	83	5	...	...	...	....

* Average figure for check plats in soil-treatment experiments.

It is evident that with colorado spruce and white pine, losses were practically uniform on beds sown either in the spring or in the fall. The same is true of the beds of norway spruce at location A. At location B, however, losses were high in the fall-sown beds but were considerably lower than in those sown on May 6 and 7. High losses were noted also at location D, but it should be stated that these beds were considerably behind the other fall-sown beds in time of emergence. Losses on the red-pine beds at location A were greatest in those sown on April 21, but were just as high on the few fall-sown beds at location D. The fact that the most severe damping-off occurred in the beds of red pine and norway spruce which were sown in May does not necessarily indicate that the time of sowing was the chief factor involved, for, as already pointed out, these beds were situated in the sections of the nursery where the disease had been most prevalent in years past.

While the information obtained includes but one season's observations, the data as a whole indicate that damping-off may occur to a serious degree in fall-sown beds. The differences in prevalence of the disease at the nursery under discussion are not shown to be definitely related to the time of sowing.

Soil treatments with chemicals

Field tests with chemicals were begun at the Ithaca nursery in 1925 and continued on a more extensive scale at the Keene nursery during the following two seasons.

The twelve beds of the Ithaca nursery were used for a single experiment. Each bed was divided into two sections 4 by 6 feet in size, thus providing for three checks and three replicates of seven different treatments. Chemicals were applied in solution, both directly after seeding and at various times for several weeks thereafter. Following seeding operations the beds were covered with burlap, which was removed as soon as emergence began and was allowed to remain on top of the half-shade frames for a

Plate II

CULTURES OF RHIZOCTONIA GROWN FOR FIVE WEEKS AT 25° C.

1, 2, and 3, Strain 77 on Czapek's, malt-extract, and potato-dextrose agars, respectively
4, 5, and 6, Strain 156 on Czapek's, malt-extract, and potato-dextrose agars, respectively
7, 8, and 9, Strain 167 on Czapek's, malt-extract, and potato-dextrose agars, respectively
10, 11, and 12, Strain 187 on Czapek's, malt-extract, and potato-dextrose agars, respectively

1
4
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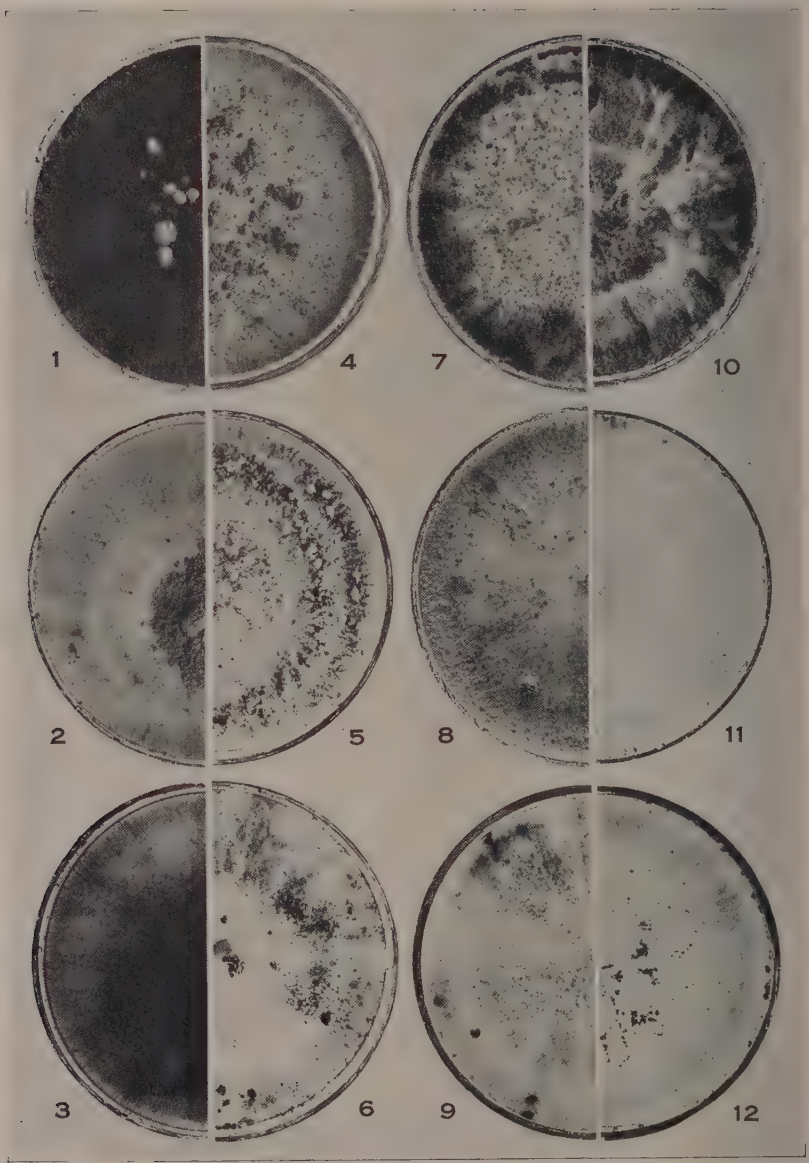
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Plate III

CULTURES OF RHIZOCTONIA GROWN FOR FIVE WEEKS AT 25° C.

1, 2, and 3, Strain 193 on Czapek's. malt-extract, and potato-dextrose agars, respectively
4, 5, and 6, Strain 183 on Czapek's, malt-extract, and potato-dextrose agars, respectively
7, 8, and 9, Strain 1 on Czapek's, malt-extract, and potato-dextrose agars, respectively
10, 11, and 12, Strain 192 on Czapek's, malt-extract, and potato-dextrose agars, respectively

period of four weeks. This additional shading, together with frequent sprinkling, maintained a rather high soil moisture and atmospheric humidity.

The experiments at the Keene nursery were conducted on an entirely different plan. Throughout both seasons it was attempted to conform in every possible way to the regular nursery practices in making up and caring for the beds. In order to provide for adequate replication of the various treatments, the beds were divided by board partitions into continuous series of small plats In the earliest experiments of 1926, these plats were 12 square feet in area and were sown with a definite quantity of seed distributed as evenly as possible in nine rows. For all later experiments the size of the plats was increased to 16 square feet, the number of rows remaining the same. The seed required for each set of three rows was weighed separately, thus providing for a more nearly uniform distribution than that afforded by seeding the entire nine rows from a single packet.

Liquid treatments were made from ordinary sprinkling cans directly after seeding. In some cases further treatments were made after emergence had occurred. Half of the check plats were sprinkled with an amount of water equal to that used in applying the chemicals to the treated plats. Where chemicals were applied in the dry condition, the materials were sifted on the plats from an envelope and worked into the upper inch of soil when the beds were given the final raking.

In the spring experiments of 1926, four replications were made of each treatment and a check plat was provided for every five to seven treated plats. In later tests, five replications were made and a single plat was left untreated for every five which were treated. The various replicates and checks were distributed over the beds as nearly uniformly as possible.

Materials

Aluminum sulfate, copper carbonate, mercuric chloride, mercurous chloride, sulfuric acid, and a number of commercial preparations containing organic compounds of mercury, were used at various times in the course of these investigations. The source and the grades of the different materials used in these experiments are given below. The descriptions of the composition and the chemical properties of the various commercial preparations are based upon statements of the manufacturers. The same is true for the changes in composition which have been made in several of the materials since the particular samples used in these tests were received.

Aluminum sulfate of technical grade was obtained from Merck & Co., of New York City, and a commercial grade was procured from the General Chemical Co., of New York City. By passing the pulverized form through sieves with twenty-five meshes to the inch, a fairly satisfactory material was obtained for use in the experiments with dry chemicals. This product was much coarser, however, than the mercury dusts.

The sulfuric acid employed was the commercial grade of concentrated acid, purchased in a local drug store.

Copper carbonate of technical grade was procured from the J. T. Baker Chemical Co., of Phillipsburg, New Jersey.

Mercuric chloride, U. S. P. IX, was obtained from Merck & Co., and mercurous chloride (calomel) from the Mallinckrodt Chemical Works, of New York City, under the trade name *Calogreen*. For the experiments with dry chemicals these two materials were diluted with talc.

The organic mercury preparations contained as their toxic ingredient either chlorophenol mercury (hydroxymercurichlorophenol sulfate), cyancresol mercury, or nitrophenol mercury (hydroxymercurinitrophenol sulfate). Of these preparations, Uspulun, Bayer compound, Bayer dust, and preparations I–8, II–6, 101, and 112, were furnished by the Bayer Company, of New York City. Their toxic constituents are as follows:

Name of preparation	Name and percentage of toxic constituents	Approximate solubility of the toxic constituents in water
		(Per cent)
Bayer compound	10% nitrophenol mercury	Complete
Bayer I–8	8% nitrophenol mercury	5
Bayer II–6	6% nitrophenol mercury	10
Bayer dust	4% nitrophenol mercury	5
Dipdust	2% nitrophenol mercury and 6% chlorophenol mercury	20
Bayer 112	3% nitrophenol mercury and 3% chlorophenol mercury	5
Bayer 101	10% chlorophenol mercury	80
Uspulun	30% chlorophenol mercury	Complete

Semesan Bel, Semesan, and Semesan Jr., obtained from E. I. du Pont de Nemours & Co., of Wilmington, Delaware, contained respectively 10 per cent, 35 per cent, and 10 per cent of chlorophenol mercury. This chemical as it occurred in Semesan was almost completely soluble in water, in Semesan Bel it was only slowly soluble, and in Semesan Jr. approximately one-third was immediately soluble.[6]

Germisan, containing approximately 31 per cent of cyancresol mercury, was obtained from the Saccharin-Fabrik, Aktiengesellschaft, Magdeburg Südost, Germany.

The mercury content of chlorophenol mercury is 61 to 62 per cent, in comparison with 55 per cent in nitrophenol mercury, 73.89 per cent in mercuric chloride, and 84.96 per cent in mercurous chloride.

[6] Several changes have been made in the composition of two of these preparations since the experiments here reported were conducted. Thus, as manufactured in June, 1928, Semesan Bel contained the chlorophenol mercury in a form which was about 40 per cent immediately soluble, while Semesan Jr. contained as the toxic constituent 12 per cent of cresol mercury, approximately 40 per cent of which was readily soluble.

Methods of taking and interpreting data

The methods employed in taking data on damping-off were varied somewhat from season to season, depending largely upon the stage of seedling development at which the disease first appeared. Thus, in the spring and early summer experiments of 1926, emergence in the beds of norway spruce and red pine was practically complete before more than a trace of damping-off had appeared. Counts of all seedlings in three rows in each plat were made on such beds as soon as emergence had taken place. Counts were again taken of the final stand in the same rows in early September. Observations made during the intervening time indicated that the differences between the two sets of figures represented losses caused by the disease. Damping-off appeared in beds of white pine soon after the first seedlings had pushed through the soil, and continued during and after the emergence period. Because of this, a single count of the final stand was made in Sepetmber, on four rows in each plat.

During the season of 1927, losses from damping-off were evident in the beds of norway spruce and red pine from the very beginning of the emergence period. This fact, combined with the loss of a number of seedlings by bird injury,[7] made frequent counts necessary. All plats were therefore inspected at short intervals to record and remove from four rows in each plat all seedlings which were damped-off and the stubs of seedlings which had been broken off by birds. As soon as emergence was over, the surviving seedlings were counted, and this was followed by another count at the end of the season. The difference between the two indicated the loss from damping-off during this period. Where losses from disease were slight throughout the season, it frequently happened that the number damped-off was more than equaled by the number of late-emerging seedlings which always appear in conifer seedbeds even many weeks after the vast majority of seedlings have emerged. These differences are indicated as plus figures under the heading of percentage of damping-off, as in the 1926 experiments. In all cases they are small and are entirely without significance.

In the beds of white pine, damping-off likewise appeared during the general emergence period. The more severe bird injury in these beds made it desirable to count only the final stand at the close of the season. It is pointed out later in this work that the results of these experiments on white pine cannot be accorded the same significance as those with red pine and norway spruce.

As a result of the unequal distribution of the losses resulting from bird injury, a sound interpretation of the data obtained from the norway-spruce and red-pine experiments of 1927 necessarily involved the introduction of a correcting factor. This was done in the following manner.

[7] When conifer seedlings first emerge from the seedbeds, the immature cotyledons still inclosed within the seed coat are borne at the tip of the hypocotyl. The type of bird injury referred to was occasioned by the action of sparrows and goldfinches in feeding on these tips. As a result, only the broken hypocotyl remained. The danger from bird injury existed only until the cotyledons were mature and the seed coats shed.

For each plat the total loss from bird injury was subtracted from the total emergence. Losses from damping-off were then based upon this remainder, which represented the total number of seedlings surviving bird injury and from which the percentage of damping-off was calculated. It was then assumed that the seedlings which were destroyed by birds would have damped-off at the same rate as those which were not destroyed. The final stand as indicated in the various tables therefore represents the total emergence minus the sum of the actual damping-off and the theoretical damping-off estimated for the seedlings removed by bird injury. The data on final stand are given on the basis of nine rows in each plat.

Essentially the same method was employed in interpreting the data from the Ithaca experiment of 1925, there having been a rather uneven occurrence of losses due to mechanical breakage.

The figures for each set of replicate plats were averaged and the probable error of this mean was determined by the use of Bessel s formula. The same was done for the check plats of each experiment. In view of the rather limited number of replications employed, differences between any two means are not considered significant unless they have at least four times the value of the probable error of this difference.

Results of the soil-treatment experiments

The results obtained from the soil treatments are presented according to the species of conifer sown in the experimental plats.

Red pine — Liquid treatments. Preliminary field tests with sulfuric acid and several mercury disinfectants were made on beds of red pine at the Ithaca nursery in 1925. The beds were artificially infested with a damping-off fungus (Rhizoctonia) and were maintained under rather wet conditions. The incidence of disease was very high (table 8). Thus in the untreated

TABLE 8. LIQUID TREATMENTS ON BEDS OF RED PINE SOWN AT THE ITHACA NURSERY ON MAY 15, 1925

Material employed	Number of applications	Amount per square foot	Results (average of three replications)		
			Percentage of emergence	Percentage of damping-off after emergence	Final stand††
Bayer compound	5	2.58 gms.*	30.0±2.1	43.3± 4.8	260±29.9
Germisan	2	1.33 gms.†	.....	.....	.....
Mercuric chloride	5	1.03 gms.‡	46.0±1.0	22.0± 3.5	536±61.2
Sulfuric acid	1	5 cc.§	36.0±3.3	61.7±10.6	230±67.3
Uspulun	2	3.01 gms.‖	39.7±1.8	24.3± 3.5	449±33.0
Uspulun	3	3.34 gms.¶	28.7±1.4	28.0± 2.1	307±17.1
Uspulun	6	4.02 gms.**	35.3±5.9	19.7± 3.6	442±85.7
Check (untreated)	.....	.....	25.7±3.9	98.0± 0.8	8± 3.1

* 0.7 gram in 400 cc. water at time of seeding; 0.47 gram in 270 cc. four, six, eight, and ten weeks later.
† 0.8 gram in 400 cc. water at time of seeding; 0.53 gram in 270 cc. four weeks later.
‡ 0.33 gram in 400 cc. water at time of seeding; 0.22 gram in 270 cc. four weeks later; 0.16 gram in 270 cc. six, eight, and ten weeks after seeding.
§ Equivalent to from 5/32 to 6/32 fluid ounce of concentrated acid dissolved in 750 cc. water and applied at time of seeding.
‖ 2.34 grams in 940 cc. water at time of seeding; 0.67 gram in 270 cc. four weeks later.
¶ 2.0 grams in 800 cc. water at time of seeding; 0.67 gram in 270 cc. four and six weeks later.
** 0.67 gram in 270 cc. water at time of seeding, and repeated two, four, six, eight, and ten weeks later.
†† A total of 1500 seeds were sown in the area of each section from which data were obtained.

beds 98 per cent of the seedlings were destroyed, the greater part of the loss occurring during the first week after emergence began. Treatments with mercuric chloride and Uspulun caused a considerable reduction in damping-off and a marked increase in final stand. So many seedlings were killed by the second application of Germisan that these beds were discarded. Bayer compound and sulfuric acid were distinctly less effective in controlling the disease than were the other chemicals. While the experiment was carried out on a small scale and under rather artificial conditions, the results indicated that several of the materials were promising for future tests.

In 1926 a more extensive experiment was conducted with this species at the Keene nursery, solutions of aluminum sulfate, sulfuric acid, and three mercury fungicides, being used. Damping-off did not appear to any appreciable extent until emergence was nearly complete.

With the exception of the two weakest applications of sulfuric acid, and treatments 12 and 18 with Bayer compound and mercuric chloride, respectively, significant differences in percentage of loss after emergence occurred in all cases (table 9).

Because of the low incidence of disease in the check plats and the variation within any group of replicates, significant differences in final stand between treated and untreated plats did not occur with treatments 2, 5, 12, 15, 17, 18, 19, and 20.

Slight stunting was noted in several plats treated with the strongest sulfuric-acid solution, and in those plats where second applications of mercuric chloride were made. However, observations made at the end of the second growing season failed to reveal any difference in the size or the appearance of the seedlings on treated and untreated plats. Evidently those seedlings surviving the second application of mercuric chloride were not permanently stunted.

The experiment showed that certain concentrations of all of the fungicides were effective in controlling damping-off, even when applied only at seeding time, and that such treatments did not cause chemical injury.

Three experiments were conducted with liquid treatments on red pine during the season of 1927. Experiment C was sown on April 21 at the rate of 2700 seeds in each plat, while experiments A and B were sown on May 12 and May 16, respectively, at the rate of 1800 seeds. The results for the three experiments are given in table 10. The percentage of emergence was in all cases somewhat higher in the treated plats. In experiments A and B, 60.3 per cent and 38.7 per cent, respectively, of the seedlings which emerged in the checks were destroyed by damping-off, while all treatments reduced the disease to a negligible amount. The final stand for all treatments of experiments A and B was very high, and in all cases was significantly greater than that of the check. The differences between the final stands of the several treatments are, however, not significant.

TABLE 9. LIQUID TREATMENTS ON BEDS OF RED PINE SOWN AT THE KEENE NURSERY ON MAY 26, 1926

Treatment no.	Material employed	Rate of application per square foot				Number of replications	Results		
		At seeding time		Two months after seeding			Percentage of emergence	Percentage of loss after emergence	Final stand per 9-row plat
		Weight of material	Quantity of water in which applied	Weight of material	Quantity of water in which applied				
			(Cubic centimeters)		(Cubic centimeters				
1	Aluminum sulfate	7.087 gms.	500	.....	.....	4	72.0 ± 3.1	6.8 ± 2.2	1,876 ± 74.6
2		10.631 gms.	500	.....	.....	4	67.0 ± 4.4	3.3 ± 0.8	1,796 ± 107.8
3		14.175 gms.	500	.....	.....	3	71.0 ± 0.7	1.3 ± 1.6	1,959 ± 20.5
4		17.718 gms.	500	.....	.....	3	72.3 ± 1.8	4.0 ± 2.2	1,943 ± 11.7
5	Sulfuric acid	2/32 fl. oz.	500	.....	.....	4	63.3 ± 2.9	14.8 ± 4.5	1,491 ± 59.2
6		3/32 fl. oz.	500	.....	.....	3	70.3 ± 2.7	10.7 ± 4.3	1,744 ± 19.9
7		4/32 fl. oz.	500	.....	.....	3	62.7 ± 1.4	7.3 ± 2.4	1,621 ± 9.1
8		5/32 fl. oz.	500	.....	.....	3	66.3 ± 1.1	1.7 ± 1.9	1,822 ± 10.0
9		6/32 fl. oz.	500	.....	.....	4	66.5 ± 2.0	2.0 ± 0.5	1,824 ± 62.6
10	Bayer compound	2.625 gms.	750	.....	.....	4	69.5 ± 2.2	7.8 ± 2.3	1,780 ± 19.5
11		1.875 gms.	750	.....	.....	4	68.0 ± 1.5	10.3 ± 1.6	1,715 ± 54.4
12		1.875 gms.	750	1.25 gms.	500	4	70.3 ± 1.6	9.8 ± 4.1	1,782 ± 111.4
13		1.250 gms.	500	.....	.....	4	72.3 ± 1.8	4.3 ± 1.0	1,941 ± 64.6
14		1.250 gms.	500	0.625 gm.	250	4	70.0 ± 2.2	4.3 ± 1.0	1,870 ± 64.7
15*	Mercuric chloride	0.6206 gm.	750	.....	.....	4	63.0 ± 1.2	6.5 ± 2.0	1,642 ± 30.6
16		0.4433 gm.	750	.....	.....	3	63.7 ± 0.8	3.0 ± 1.4	1,726 ± 13.8
17		0.4433 gm.	750	0.2955 gm.	500	3	66.3 ± 2.6	10.3 ± 1.8	1,667 ± 54.1
18		0.2955 gm.	500	.....	.....	3	69.7 ± 2.9	11.7 ± 5.4	1,744 ± 171.7
19		0.2955 gm.	500	0.1478 gm.	250	4	67.5 ± 1.0	12.0 ± 3.1	1,669 ± 70.5
20*	Uspulun	2.625 gms.	750	.....	.....	3	69.0 ± 2.9	8.7 ± 0.2	1,760 ± 80.9
21		1.875 gms.	750	.....	.....	3	68.7 ± 0.2	5.0 ± 1.8	1,826 ± 31.0
22		1.875 gms.	750	1.250 gms.	500	4	69.5 ± 2.0	8.5 ± 2.7	1,786 ± 82.9
23		1.250 gms.	500	.....	.....	4	70.3 ± 1.4	6.0 ± 2.5	1,856 ± 74.1
24		1.250 gms.	500	0.625 gm.	250	4	76.0 ± 2.2	8.5 ± 2.4	1,948 ± 55.4
	Check (untreated)	.....	.....	.....	.....	12	69.9 ± 1.4	30.8 ± 3.5	1,350 ± 67.4

* Treatments 15 and 20 are equivalent in mercury content, as are also treatments 16 and 21, 17 and 22, 18 and 23, and 19 and 24.

TABLE 10. LIQUID TREATMENTS ON BEDS OF RED PINE AT THE KEENE NURSERY, SEASON OF 1927

	Treatment no.	Treatment: Material employed	Treatment: Amount per square foot applied in 500 cc. of water	Number of replications	Results: Percentage of emergence	Results: Percentage of damping-off after emergence	Results: Final stand per 9-row plat
Experiment A	1	Aluminum sulfate	10.631 gms.	5	81.2 ± 1.8	6.8 ± 2.9	1,369 ± 61.0
	2		14.175 gms.	4	79.8 ± 1.5	1.5 ± 0.6	1,413 ± 33.1
	3		17.718 gms.	5	77.4 ± 1.9	1.0 ± 0.7	1,367 ± 35.4
	4	Sulfuric acid	4/32 fl. oz.	5	79.2 ± 1.1	7.8 ± 2.0	1,313 ± 42.8
	5		5/32 fl. oz.	5	74.2 ± 1.1	8.0 ± 2.4	1,231 ± 31.3
	6		6/32 fl. oz.	5	78.2 ± 2.4	3.4 ± 0.3	1,351 ± 40.9
	7*	Mercuric chloride	0.1182 gm.	6	76.0 ± 1.9	0.3 ± 1.5	1,337 ± 34.2
	8		0.2069 gm.	6	73.7 ± 2.6	+1.0 ± 1.3	1,312 ± 44.7
	9	Semesan	0.500 gm.	6	75.3 ± 2.8	3.2 ± 2.7	1,292 ± 49.9
	10		0.875 gm.	6	77.0 ± 2.6	0.8 ± 1.0	1,355 ± 43.9
	11*	Uspulun	0.500 gm.	6	76.2 ± 3.0	4.8 ± 3.2	1,305 ± 82.0
	12		0.875 gm.	6	74.2 ± 1.8	+1.3 ± 1.0	1,325 ± 34.5
		Check (untreated)	.....	15	64.5 ± 2.1	60.3 ± 5.5	514 ± 77.6
Experiment B	1	Mercuric chloride	0.2364 gm.	6	74.0 ± 1.8	+0.2 ± 0.4	1,321 ± 32.6
	2		0.4137 gm.	6	71.8 ± 1.2	+0.3 ± 0.3	1,292 ± 22.3
	3	Semesan	1.00 gm.	6	73.7 ± 2.5	+0.3 ± 1.3	1,303 ± 40.2
	4		1.75 gms.	6	70.8 ± 2.3	2.2 ± 1.3	1,234 ± 27.1
	5	Uspulun	1.00 gm.	5	75.8 ± 1.4	0.3 ± 0.8	1,351 ± 24.2
	6		1.75 gms.	6	71.8 ± 1.1	+0.7 ± 0.6	1,283 ± 22.5
		Check (untreated)	.....	7	66.9 ± 2.9	38.7 ± 7.1	780 ± 106.8
Experiment C	1	Aluminum sulfate	10.631 gms.	3	66.7 ± 2.1	2.7 ± 1.2	1,739 ± 35.5
	2		17.718 gms.	3	64.7 ± 2.4	6.7 ± 3.0	1,618 ± 55.8
	3	Sulfuric acid	4/32 fl. oz.	3	58.0 ± 3.4	20.3 ± 11.1	1,203 ± 120.9
	4		6/32 fl. oz.	3	60.3 ± 0.9	11.7 ± 2.6	1,445 ± 24.3
		Check (untreated)	.....	5	55.2 ± 4.6	48.0 ± 4.4	811 ± 127.4

* Treatments 7 and 11, and 8 and 12, of experiment A are equivalent in mercury content, as are also treatments 1 and 5, and 2 and 6, of experiment B.

It is apparent, then, that the liquid treatments were equally effective in controlling damping-off, even where very small amounts of the mercury materials were used and under conditions in which severe damping-off occurred in the check plats (figure 2). With the exception of an occasional

FIGURE 2. FINAL STAND OF RED-PINE SEEDLINGS IN PLATS TREATED WITH LIQUID APPLICATIONS OF CHEMICALS

The numbers below the columns refer to the treatment numbers given in table 10

group of slightly stunted seedlings in several plats treated with the stronger applications, no evidence of chemical injury could be detected (Plate IV, 1 and 2, and Plate V, 1).

In experiment C, sulfuric acid was less effective than aluminum sulfate in controlling the disease. The results with the weaker acid treatment were not significantly different from those obtained from the check plats.

Red pine — Treatments with dry chemicals. Preliminary experiments were conducted at the Keene nursery in 1926 with applications of finely divided copper carbonate and Bayer dusts I–8, 101, and 112, at the rate of 4 and 8 grams per square foot. Favorable results were obtained on a small scale with the mercury dust, while the copper carbonate caused severe injury. Bayer dust, Dipdust, and copper carbonate were similarly used on fall-sown beds of the same year. The copper carbonate (1, 2, and 3 grams per square foot) did not control the disease in the smaller amounts and caused injury in the larger applications. The mercury dusts (3 and 6 grams per square foot) were only partially effective against the disease.

Spring applications of dusts made directly on the surface of fall-sown plats just prior to emergence, were approximately as effective as the fall treatments. Dusting the seed alone was practically without effect.

An experiment was conducted on red pine during the season of 1927 with dry applications of aluminum sulfate and a number of organic and inorganic mercury materials (table 11). The experiment was comparable to experiment A of the liquid treatments of the same season.

Many of the mercury treatments were effective in reducing the disease from 67.7 per cent, as noted for the checks, to an almost negligible amount. From a consideration of the percentage of damping-off and the final stand (figure 3), it would appear that the best results were obtained with the

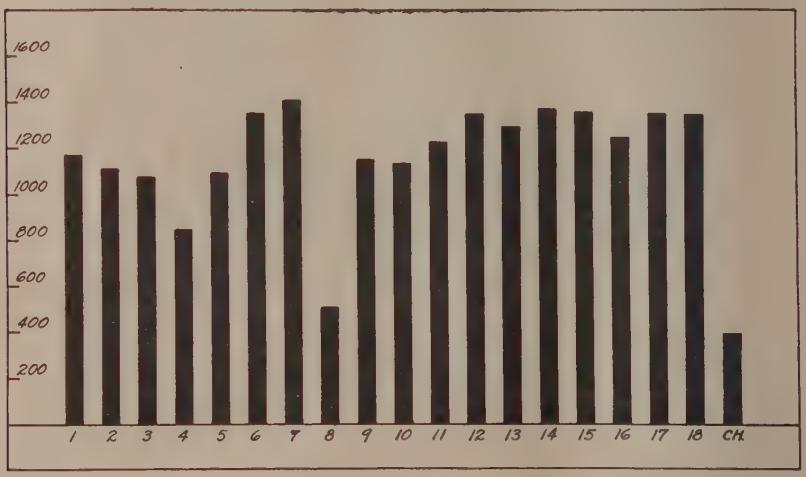


FIGURE 3. FINAL STAND OF RED-PINE SEEDLINGS IN PLATS TREATED WITH DRY APPLICATIONS OF CHEMICALS

The numbers below the columns refer to the treatment numbers given in table 11

two treatments with Bayer II-6, the heavier application of mercuric chloride, and the two heaviest applications of mercurous chloride and of Semesan Jr. The 2-gram applications of Bayer dust and of Dipdust were, on the other hand, rather ineffective in controlling the disease. Of the remaining treatments, those with the two heaviest applications of aluminum sulfate, the 4-gram application of Bayer dust, and the 4- and 6-gram applications of Dipdust, gave the least satisfactory results (Plate V, 2).

No evidence of chemical injury was noted except in a few of the plats receiving treatments 11, 12, 14, 15, and 18, where there was an indication

TABLE 11. TREATMENTS WITH DRY CHEMICALS ON BEDS OF RED PINE AT THE KEENE NURSERY, SEASON OF 1927

Treatment no.	Treatment		Number of replications	Results		
	Material employed	Amount applied per square foot		Percentage of emergence‡	Percentage of damping-off after emergence	Final stand per 9-row plat
1	Aluminum sulfate	7.087 gms.	5	72.8 ± 1.4	10.8 ± 2.3	1,168 ± 51.3
2		10.631 gms.	5	71.0 ± 2.7	13.2 ± 1.7	1,110 ± 56.7
3		14.175 gms.	4	73.0 ± 1.7	17.0 ± 5.5	1,079 ± 48.6
4	Bayer dust	2 gms.	5	70.8 ± 2.8	33.2 ± 3.6	846 ± 43.5
5		4 gms.	5	78.6 ± 2.4	23.2 ± 3.6	1,099 ± 79.7
6	Bayer II–6	4 gms.	4	77.5 ± 0.8	3.3 ± 0.7	1,355 ± 12.3
7		6 gms.	5	80.4 ± 2.9	2.4 ± 0.7	1,410 ± 51.8
8	Dipdust	2 gms.	4	65.8 ± 5.4	58.8 ± 6.5	510 ± 104.4
9		4 gms.	5	81.4 ± 2.5	22.2 ± 4.4	1,153 ± 91.1
10		6 gms.	5	71.0 ± 1.6	11.6 ± 1.9	1,131 ± 41.7
11*	Mercuric chloride	0.2364 gm.	5	72.6 ± 2.2	6.4 ± 1.7	1,228 ± 50.1
12		0.4137 gm.	4	78.5 ± 2.5	4.0 ± 1.2	1,351 ± 37.6
13*	Mercurous chloride	0.2056 gm.	5	75.2 ± 0.8	4.0 ± 1.4	1,297 ± 25.7
14		0.3598 gm.	5	79.2 ± 1.4	3.2 ± 1.0	1,376 ± 28.8
15		0.3598 gm.†	5	76.2 ± 3.1	0.6 ± 0.5	1,360 ± 53.1
16	Semesan Jr.	2 gms.	5	80.8 ± 3.1	13.8 ± 4.1	1,246 ± 73.4
17		4 gms.	5	79.0 ± 1.3	4.6 ± 1.3	1,354 ± 30.3
18		6 gms.	5	76.8 ± 0.7	2.6 ± 0.6	1,345 ± 13.7
	Check (untreated)	.....	19	59.6 ± 2.1	67.7 ± 3.6	397 ± 51.9

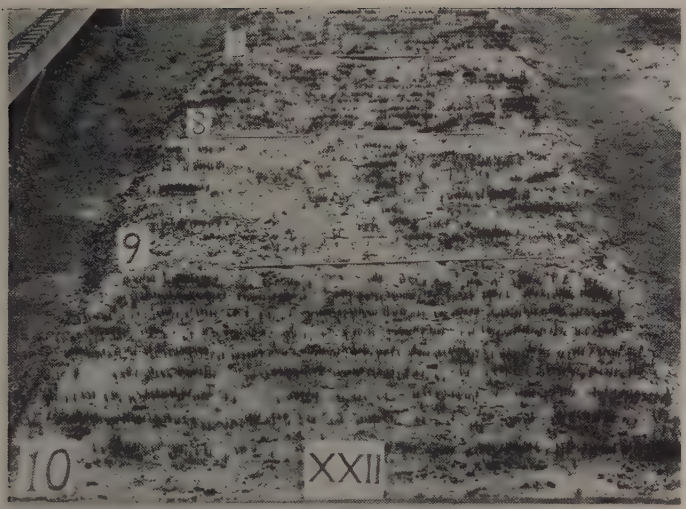
* Treatments 11 and 13 are equivalent in mercury content, as are also treatments 12, 14, and 15.
† Applied in 250 cc. water immediately after seeding.
‡ Sown at the rate of 1800 seeds in each plat.

Plate IV

FINAL STAND OF RED-PINE SEEDLINGS IN BEDS RECEIVING LIQUID TREATMENTS FOR CONTROL OF DAMPING-OFF

1: Plat 9, untreated; plat 7, 0.875 gram of Semesan per square foot; plat 8, 0.1182 gram of mercuric chloride per square foot; plat 10, 0.2069 gram of mercuric chloride per square foot

2: Plat 15, untreated; plat 13, 0.875 gram of Uspulun per square foot; plat 14, 0.500 gram of Semesan per square foot; plat 16, 4/32 fluid ounce of sulfuric acid per square foot



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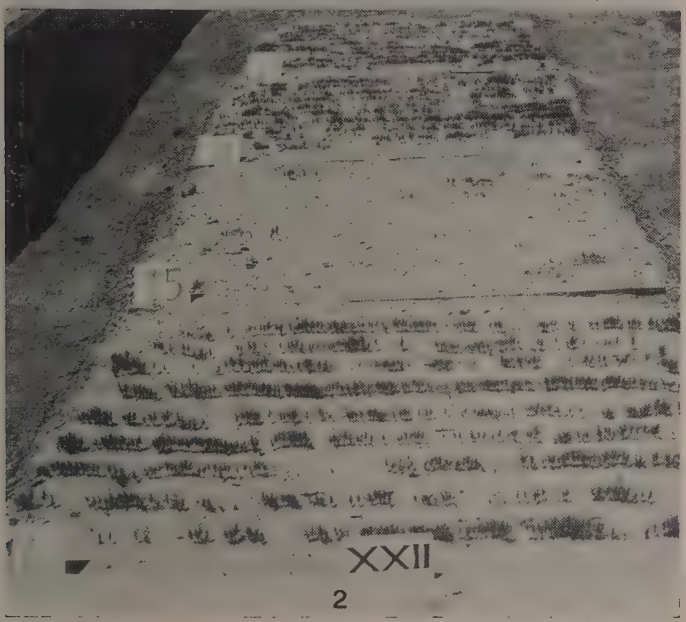

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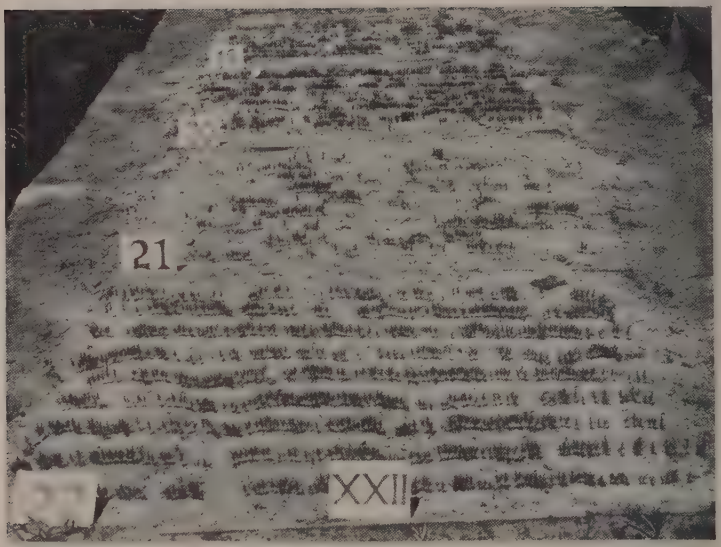

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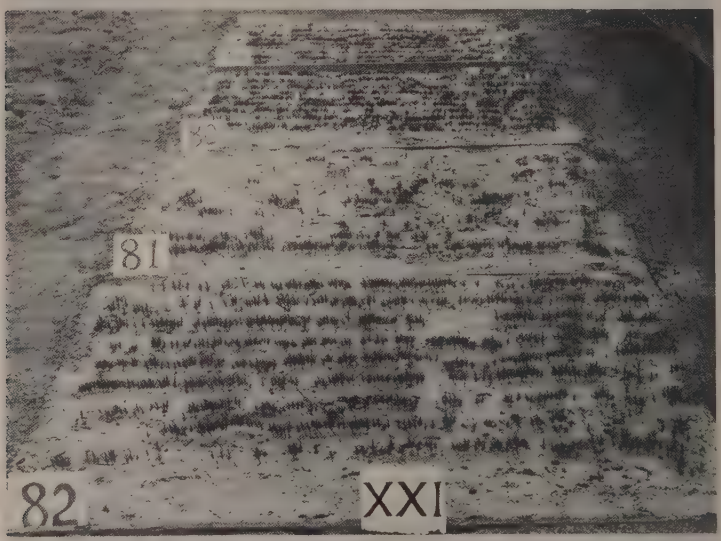

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Plate V

FINAL STAND OF RED-PINE SEEDLINGS IN BEDS TREATED WITH CHEMICALS FOR CONTROL OF DAMPING-OFF

1: Liquid treatments: Plat 21, untreated; plat 19, 3/8 ounce of aluminum sulfate per square foot; plat 20, 4/8 ounce of aluminum sulfate per square foot; plat 22, 0.875 gram of uspulun per square foot

2: Treatments with dry chemicals: Plat 81, untreated; plat 79, 0.4137 gram of mercuric chloride per square foot; plat 80, 0.2056 gram of mercurous chloride per square foot; plat 82, 0.3598 gram of mercurous chloride per square foot

of very slight stunting. This injury is of no particular significance except to indicate the maximum amount of these chemicals which can safely be employed under these conditions.

Norway spruce — Liquid treatments. Solutions of mercuric chloride and several organic mercury materials were used on spring-sown beds of norway spruce during the season of 1926. The details of the experiment are recorded in table 12. The low figures for percentage of emergence in both the treated and the untreated plats are due in part to the low viability of the seed and in part to the fact that the seeds were sown at a greater depth than usual. Damping-off, which occurred chiefly in the check plats, was rather unevenly distributed. The heavy losses noted from treatments 2, 8, 10, and 12 were due primarily to the injurious effects of the second application of chemical. Although the losses with the other treatments were considerably lower than those of the check, significant differences occurred only with treatments 1, 3, 6, 13, 15, 17, 21, and 23. Similarly, for the final stand significant differences were found only with treatments 11, 15, and 23.

Second applications of mercuric chloride and of the stronger treatment with Bayer compound caused marked injury. This was manifested by a killing of some seedlings and a severe stunting of others. The larger amounts of the other chemicals caused slight retardation, while with the smaller amounts slight stunting was noted in several of the replicates. With the single-application treatments, retardation was noted in several plats where the larger amounts were used. Observations were again made at the end of the second growing season. Although considerable variation in the size of the seedlings occurred, it was possible to group the plats in a general way. In the plats of class A, therefore, most of the seedlings were typical of the average stock commonly used for transplanting. In those of class B the stock was of a somewhat smaller size than in A, but large numbers of these seedlings would normally be transplanted. In those of class C the majority of the seedlings were of decidedly inferior quality and would be discarded.

This experiment indicates that norway spruce is rather susceptible to chemical injury, and that of the treatments employed only the single applications with the smaller amounts are practicable for use on this species under these conditions.

Liquid treatments were used also in one series of norway-spruce plats sown in the fall of 1926. So little damping-off occurred that counts were made only of the final stand. From table 13 it is seen that stunting occurred from a number of the treatments. Since germination did not take place until the spring of 1927, five months after the time of treating, it would appear that either the chemicals exerted a toxic action upon the dormant seed, or else they were present in a readily available state at the time of germination. The latter hypothesis seems the more tenable.

TABLE 12. LIQUID TREATMENTS ON BEDS OF NORWAY SPRUCE SOWN AT THE KEENE NURSERY ON MAY 21, 1926

Treatment no.	Material employed	Treatment: Rate of application per square foot: At seeding time: Weight of material	At seeding time: Quantity of water in which applied	On July 8: Weight of material	On July 8: Quantity of water in which applied	Number of replications	Results: Percentage of emergence†	Percentage of loss after emergence	Final stand per 9-row plat	Classification of plats
			(Cubic centimeters)		(Cubic centimeters)					
1	Bayer compound	1.250 gms.	500	0.625 gm.	250	4	53.5 ± 0.2	+1.3 ± 2.9	1,680 ± 42.3	A
2		1.750 gms.	500	0.875 gm.	250	4	52.8 ± 1.5	9.3 ± 2.9	1,478 ± 22.9	A
3		1.875 gms.	750	.....	.....	4	43.5 ± 4.8	+2.8 ± 3.3	1,387 ± 170.8	A
4		2.625 gms.	750	.....	.....	4	50.0 ± 1.8	3.5 ± 3.4	1,496 ± 79.0	A
5*	Mercuric chloride	0.2955 gm.	500	.....	.....	4	53.5 ± 1.5	2.8 ± 2.6	1,607 ± 26.5	A
6		0.2955 gm.	500	0.1478 gm.	250	4	54.8 ± 1.0	2.8 ± 0.7	1,636 ± 33.3	A
7		0.4137 gm.	500	.....	.....	3	53.8 ± 2.0	4.0 ± 1.5	1,533 ± 60.2	A
8		0.4137 gm.	500	0.2069 gm.	250	4	56.0 ± 2.7	21.3 ± 2.0	1,382 ± 69.5	C
9		0.4433 gm.	750	.....	.....	4	56.5 ± 2.6	7.5 ± 2.1	1,606 ± 59.2	A
10		0.4433 gm.	750	0.2955 gm.	500	4	56.0 ± 1.1	11.0 ± 3.0	1,551 ± 74.8	B
11		0.6206 gm.	750	.....	.....	3	54.5 ± 2.1	1.3 ± 2.8	1,720 ± 41.7	B
12		0.6206 gm.	750	0.4137 gm.	250	3	53.0 ± 0.5	21.0 ± 3.3	1,312 ± 59.4	C
13	Semesan	1.250 gms.	500	0.625 gm.	250	4	58.0 ± 2.6	+2.8 ± 2.8	1,855 ± 115.5	A
14		1.750 gms.	500	0.875 gm.	250	3	56.3 ± 0.7	4.3 ± 2.0	1,658 ± 54.9	A
15		1.875 gms.	750	.....	.....	4	55.8 ± 0.7	+4.8 ± 4.7	1,805 ± 58.2	A
16		2.625 gms.	750	.....	.....	4	52.0 ± 1.1	3.8 ± 1.8	1,545 ± 16.9	B
17*	Uspulun	1.250 gms.	500	.....	.....	4	52.8 ± 0.7	0.8 ± 0.9	1,626 ± 34.2	A
18		1.250 gms.	500	0.625 gm.	250	4	50.0 ± 4.4	4.8 ± 1.9	1,477 ± 128.2	A
19		1.750 gms.	500	.....	.....	4	53.3 ± 2.2	2.5 ± 1.8	1,612 ± 59.3	B
20		1.750 gms.	500	0.875 gm.	250	4	55.3 ± 2.6	4.0 ± 2.2	1,650 ± 86.6	B
21		1.875 gms.	750	.....	.....	4	51.5 ± 2.4	0.8 ± 1.3	1,582 ± 58.8	B
22		1.875 gms.	750	1.250 gms.	500	4	50.5 ± 2.9	4.0 ± 4.0	1,500 ± 91.7	B
23		2.625 gms.	750	.....	.....	4	56.5 ± 0.6	+2.3 ± 2.2	1,784 ± 49.1	B
24		2.625 gms.	750	1.750 gms.	500	4	51.0 ± 2.1	3.0 ± 3.5	1,516 ± 51.7	C
	Check (untreated)	.....	.....	.....	.....	18	54.2 ± 1.2	23.1 ± 5.0	1,308 ± 85.1	A

* Treatments 5 and 17 are equivalent in mercury content, as are also treatments 6 and 18, 7 and 19, 8 and 20, 9 and 21, 10 and 22, 11 and 23, and 12 and 24.

† Based on 3100 seeds sown in each plat.

TABLE 13. LIQUID TREATMENTS ON BEDS OF NORWAY SPRUCE SOWN AT THE KEENE NURSERY ON NOVEMBER 4, 1926

Treatment no.	Treatment		Number of replications	Results	
	Material employed	Amount per square foot applied in 500 cubic centimeters of water		Final stand per 9-row plat	Condition of seedlings at end of first growing season
1	Aluminum sulfate*	3.544 gms.	5	588±27.0	Normal
2		7.087 gms.	5	608±39.0	Slightly stunted in *three* plats
3	Bayer compound	0.750 gm.	5	715±30.4	Normal
4		1.500 gms.	5	719±24.8	Normal
5	Mercuric chloride	0.1773 gm.	5	686±33.0	Slightly stunted in *one* plat
6		0.3546 gm.	5	727±26.4	Severely stunted in *all* plats
7	Semesan	0.750 gm.	5	653±30.4	Slightly stunted in *one* plat
8		1.500 gms.	5	646±45.8	Severely stunted in *all* plats
9	Sulfuric acid*	3/64 fl. oz.	5	518±36.0	Normal
10		5/64 fl. oz.	5	527±19.7	Slightly stunted in *three* plats
11	Uspulun	0.750 gm.	5	559±20.2	Slightly stunted in *one* plat
12		1.500 gms.	5	427±28.8	Severely stunted in *all* plats
	Check (untreated)	.....	12	587±24.5	Normal

* Due to an error in calculation, the aluminum sulfate and the sulfuric acid were used at half the strength originally planned.

It is rather surprising to note that the Bayer compound, shown by greenhouse experiments[8] to be more toxic than equal concentrations of Uspulun, was in this case non-injurious.

The results with liquid treatments on spring-sown beds of 1927 are presented in table 14. The plats of experiment B were sown on April 21, and those of experiment A on May 7, both at the rate of 3150 seeds. The two series were located several hundred yards apart.

In experiment A, all of the treatments caused a considerable reduction in the amount of damping-off and thereby increased the total emergence and final stand. The low percentage of emergence noted for all the plats is due to the fact that approximately only 25 per cent of the seed were viable. The three mercury materials gave the greatest reduction in percentage of damping-off. However, the stronger applications of these materials caused decided chemical injury. This was manifested by a failure of the seedlings to continue top growth. In normal seedlings the main axis elongated from one-half to three-fourths of an inch beyond the origin of the first leaves, while in severely stunted plants only one-fourth inch of such stem growth had occurred. It is doubtful whether the slight injury noted for the weaker mercury treatments is of any practical importance.

[8] Unpublished results obtained by the writer.

TABLE 14. LIQUID TREATMENTS ON SPRING-SOWN BEDS OF NORWAY SPRUCE AT THE KEENE NURSERY, SEASON OF 1927

	Treatment no.	Treatment: Material employed	Treatment: Amount per square foot applied in 500 cubic centimeters of water	Number of replications	Results: Percentage of emergence	Results: Percentage of damping-off after emergence	Results: Final stand per 9-row plat	Results: Chemical injury
Experiment A	1	Aluminum sulfate	7.087 gms.	5	23.6 ± 0.7	13.2 ± 1.1	656 ± 25.3	None
	2		10.631 gms.	5	23.2 ± 0.7	9.8 ± 1.5	662 ± 29.5	None
	3		14.175 gms.	5	24.0 ± 0.6	13.4 ± 1.7	654 ± 19.2	None
	4*	Mercuric chloride	0.1773 gm.	5	23.0 ± 0.7	6.8 ± 1.1	674 ± 24.3	Slight
	5		0.3546 gm.	5	25.0 ± 1.0	7.0 ± 0.4	729 ± 28.0	Medium
	6	Semesan	0.750 gm.	5	18.2 ± 0.5	3.8 ± 1.3	547 ± 15.7	Slight
	7		1.500 gms.	5	19.0 ± 1.0	5.0 ± 0.5	570 ± 29.2	Medium to severe
	8	Sulfuric acid	3/32 fl. oz.	5	20.8 ± 0.4	17.0 ± 2.6	546 ± 27.5	None
	9		4/32 fl. oz.	5	20.8 ± 0.8	12.6 ± 2.7	577 ± 37.8	None
	10		5/32 fl. oz.	5	20.2 ± 0.5	11.2 ± 1.7	561 ± 19.4	None
	11*†	Uspulun	0.750 gm.	5	17.4 ± 0.5	7.2 ± 1.2	513 ± 16.3	Slight
	12		1.500 gms.	3	14.3 ± 0.2	6.3 ± 2.6	416 ± 13.8	Severe
		Check (untreated)	.....	12	11.2 ± 0.7	58.4 ± 2.6	160 ± 15.3	.....
Experiment B	1	Aluminum sulfate	7.087 gms.	3	27.3 ± 1.2	10.7 ± 1.3	779 ± 41.1	None
	2		14.175 gms.	3	29.3 ± 0.6	4.3 ± 1.1	880 ± 28.4	None
	3	Sulfuric acid	3/32 fl. oz.	3	26.7 ± 1.3	5.7 ± 0.8	785 ± 44.7	None
	4		5/32 fl. oz.	3	26.0 ± 0.4	10.0 ± 0.7	736 ± 19.7	None
		Check (untreated)	.....	4	20.8 ± 2.9	36.5 ± 6.8	454 ± 9.8	.....

* Treatments 4 and 11 of experiment A are equivalent in mercury content, as are also treatments 5 and 12.
† The plats of this treatment are not so nearly uniformly distributed as are those of other treatments.

Eliminating, then, those treatments which caused appreciable injury, it is of interest to note the differences that occurred in the final stand of healthy seedlings (figure 4). In general, plats treated with mercuric

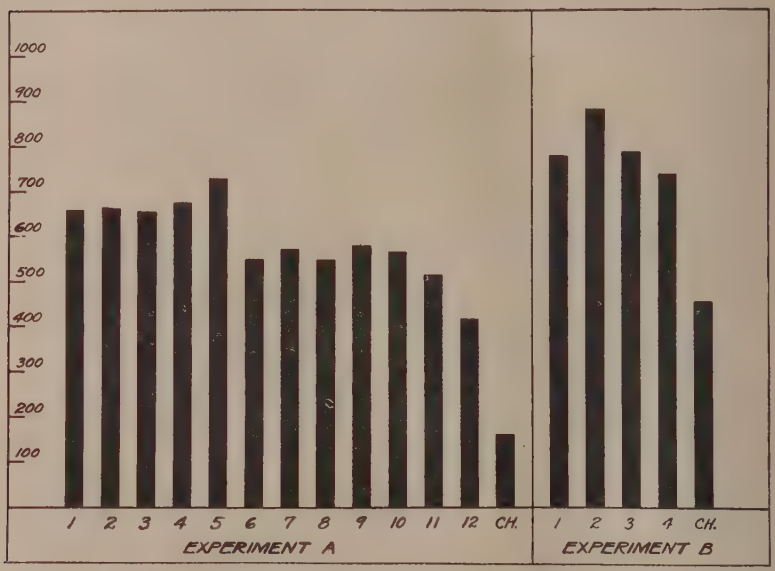


FIGURE 4. FINAL STAND OF NORWAY-SPRUCE SEEDLINGS IN PLATS TREATED WITH LIQUID APPLICATIONS OF CHEMICALS

The numbers below the columns refer to the treatment numbers given in table 14

chloride and aluminum sulfate yielded the greatest number, and those with Uspulun the lowest, while sulfuric acid and Semesan ranged between these two.

The results of experiment B indicate that both aluminum sulfate and sulfuric acid were effective in considerably reducing the incidence of disease.

Norway spruce — Treatments with dry chemicals. Copper carbonate and several organic mercury dusts were used in seed and soil treatments on both spring- and fall-sown beds of norway spruce in 1926. The results were similar to those on red pine, which was included in the same plats. Since these tests have already been discussed under red pine, they are not considered in detail here. It will be recalled, however, that soil applications of the mercury dusts gave rather promising results on the spring-sown beds.

During the season of 1927, an extensive experiment was conducted with applications of aluminum sulfate and both organic and inorganic mercury materials, on beds of this species. The experimental plats were located adjacent to those of the liquid treatments on spruce of that season, with which they are comparable.

The high incidence of disease in the check plats (table 15) is evidenced not only by the high percentage of damping-off after emergence, but also by the low percentage of emergence. With the exception of the smaller applications of several materials, all of the treatments reduced the disease to a negligible amount (Plate VI, 1 and 2).

The larger applications of mercuric chloride and mercurous chloride caused medium to severe stunting, similar to that described for several of the liquid treatments on this species. The very slight stunting noted from the smaller applications of these materials is of no practical importance.

From the data on final stand, it is evident that the two largest applications of aluminum sulfate, the two treatments with Bayer II 6, the larger applications of Bayer dust, Dipdust, and Semesan Bel, and the smaller applications of mercuric chloride and mercurous chloride, were practically equally effective (figure 5).

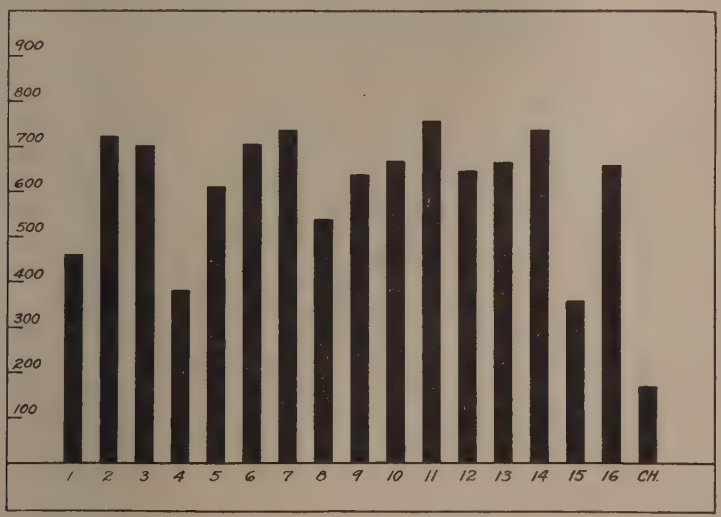


FIGURE 5. FINAL STAND OF NORWAY-SPRUCE SEEDLINGS IN PLATS TREATED WITH DRY APPLICATIONS OF CHEMICALS

The numbers below the columns refer to the treatment numbers given in table 15

TABLE 15. TREATMENTS WITH DRY CHEMICALS ON BEDS OF NORWAY SPRUCE SOWN AT THE KEENE NURSERY ON MAY 6, 1927

Treatment no.	Treatment		Number of replications	Results			
	Material employed	Amount applied per square foot		Percentage of emergence ‡	Percentage of damping-off after emergence	Final stand per 9-row plat	Chemical injury
1	Aluminum sulfate	3.544 gms.	5	18.8 ± 1.5	24.0 ± 3.5	460 ± 52.2	None
2		7.087 gms.	5	27.2 ± 1.2	14.4 ± 2.6	722 ± 22.5	None
3		10.631 gms.	5	25.2 ± 1.5	10.8 ± 2.5	701 ± 40.4	None
4	Bayer dust	2 gms.	5	18.6 ± 0.7	34.6 ± 2.8	385 ± 26.7	None
5		4 gms.	5	23.4 ± 1.2	18.4 ± 2.5	611 ± 46.3	None
6	Bayer II–6	4 gms.	5	24.2 ± 1.0	6.8 ± 1.7	705 ± 33.4	None
7		6 gms.	5	26.4 ± 1.8	11.2 ± 2.6	737 ± 51.1	None
8	Dipdust	2 gms.	5	21.0 ± 0.4	19.0 ± 2.4	539 ± 26.2	None
9		4 gms.	5	22.3 ± 1.3	9.5 ± 1.4	637 ± 41.8	None
10*	Mercuric chloride	0.1773 gm.	5	23.2 ± 1.1	8.2 ± 1.8	669 ± 33.7	Very slight
11		0.3546 gm.	4	25.0 ± 1.3	2.5 ± 2.2	757 ± 31.5	Medium to severe
12*	Mercurous chloride	0.1542 gm.	5	22.0 ± 1.2	7.0 ± 1.8	647 ± 44.7	Very slight
13		0.3084 gm.	5	21.4 ± 1.2	2.0 ± 0.8	663 ± 37.6	Medium to severe
14		0.3084 gm.†	5	23.8 ± 0.9	6.8 ± 1.3	737 ± 14.8	Medium to severe
15	Semesan Bel	2 gms.	4	16.0 ± 1.6	29.5 ± 2.3	357 ± 41.7	None
16		4 gms.	4	25.3 ± 1.4	16.8 ± 2.7	658 ± 48.1	None
	Check (untreated)	.....	17	11.6 ± 0.7	57.2 ± 2.8	168 ± 19.6	

* Treatments 10 and 12 are equivalent in mercury content, as are also treatments 11, 13, and 14.
† Applied in 250 cc. water immediately after seeding.
‡ Based on 3150 seeds sown in each plat.

White pine — Liquid treatments. An experiment was conducted with liquid treatments of several mercury materials on beds of white pine at the Keene nursery in 1926. Seed were sown on May 20 at the rate of 3000 to each plat. Since only about 25 per cent of the seed were viable, the percentage of emergence was low. Field observations showed no evidence of chemical injury at the time of emergence. Severe damping-off occurred in the check plats, while practically none was apparent in the treated plats. The data on final stand presented in table 16 demonstrate that nearly all of the treatments were effective in producing from 200 to 400 more seedlings than were recorded for the check plats. The increases were for the most part easily significant.

It is interesting to note that in nine out of ten cases in which second applications were made, the final stand was lower than for the corresponding plats receiving but a single application. Field observations showed that many seedlings were killed or stunted by the second applications of mercuric chloride, while slight stunting occurred from second applications of the other materials. Observations were repeated at the end of the second growing season, and the plats were classified on the basis of the size of seedlings contained in them. In class A are included those plats on which the majority of the seedlings were of very excellent stock, in class B those on which the seedlings were of fair size, and in class C those on which the majority of the seedlings were of decidedly inferior quality and would therefore normally be discarded. The average stock used for transplanting at the Keene nursery ranges within the classes A and B, and is designated by the combination A-B. Thus from the last column of table 16 it can be seen that the smaller amounts of the materials, particularly where single applications were made, gave the most promising results.

TABLE 16. LIQUID TREATMENTS ON BEDS OF WHITE PINE SOWN AT THE KEENE NURSERY ON MAY 20, 1926

Treatment no.	Material employed	Rate of application per square foot				Number of replications	Results	
		At seeding time		Nine weeks later			Final stand per 9-row plat	Classification of plats
		Weight of material	Quantity of water in which applied	Weight of material	Quantity of water in which applied			
			(Cubic centimeters)		(Cubic centimeters)			
1	Bayer compound	1.250 gms.	500	.....	.....	4	691 ± 31.7	A-B
2		1.250 gms.	500	0.625 gm.	250	4	613 ± 32.0	A-B
3		1.875 gms.	750	.....	.....	4	710 ± 28.9	A-B
4		1.875 gms.	750	1.250 gms.	500	4	604 ± 40.0	B-C
5		2.500 gms.	1,000	.....	.....	4	650 ± 42.2	A-B
6*	Mercuric chloride	0.4137 gm.	500	.....	.....	4	695 ± 29.1	A-B
7		0.4137 gm.	500	0.2068 gm.	250	4	645 ± 42.2	B-C
8		0.6206 gm.	750	.....	.....	4	734 ± 18.0	A-B
9		0.6206 gm.	750	0.414 gm.	500	4	629 ± 23.0	B-C
10		0.8274 gm.	1,000	.....	.....	4	716 ± 22.7	A-B
11	Semesan	1.250 gms.	500	.....	.....	4	816 ± 27.9	A-B
12		1.250 gms.	500	0.625 gm.	250	4	707 ± 50.1	B
13		1.875 gms.	750	.....	.....	4	710 ± 16.1	B
14		1.875 gms.	750	1.250 gms.	500	4	789 ± 24.3	B-C
15		2.500 gms.	1,000	.....	.....	4	761 ± 44.7	B
16	Uspulun	1.250 gms.	500	.....	.....	4	727 ± 41.2	A-B
17		1.250 gms.	500	0.625 gm.	250	4	668 ± 37.6	B
18*		1.750 gms.	500	.....	.....	4	772 ± 27.6	B
19		1.750 gms.	500	0.875 gm.	250	4	704 ± 69.9	B-C
20		1.875 gms.	750	.....	.....	4	824 ± 27.5	A-B
21		1.875 gms.	750	1.250 gms.	500	4	783 ± 23.1	B
22		2.625 gms.	750	.....	.....	4	763 ± 06.5	B
23		2.625 gms.	750	1.750 gms.	500	4	713 ± 37.4	B-C
24		2.500 gms.	1,000	.....	.....	4	603 ± 49.3	B
25		3.500 gms.	1,000	.....	.....	4	767 ± 37.7	B
	Check (untreated)	.....	.....	.....	.....	20	412 ± 31.8	A-B

* Treatments 6 and 18 are equivalent in mercury content, as are also treatments 7 and 19, 8 and 22, 9 and 23, and 10 and 25.

The results obtained with liquid treatments on this species during the season of 1927 are recorded in table 17. The plats of experiment A were

TABLE 17. LIQUID TREATMENTS ON BEDS OF WHITE PINE AT THE KEENE NURSERY, SEASON OF 1927

	Treatment no.	Treatment: Material employed	Treatment: Amount per square foot applied in 500 cubic centimeters of water	Number of replications	Final stand per 9-row plat
Experiment A	1	Aluminum sulfate	10.631 gms.	3	491 ± 9.9
	2		17.718 gms.	3	452 ± 4.5
	3	Sulfuric acid	4/32 fl. oz.	3	382 ± 29.7
	4		6/32 fl. oz.	3	365 ± 36.3
		Check (untreated)	.....	6	192 ± 39.4
Experiment B	1	Aluminum sulfate	10.631 gms.	5	318 ± 23.2
	2		14.175 gms.	5	365 ± 17.5
	3		17.718 gms.	5	397 ± 28.5
	4*	Mercuric chloride	0.1182 gm.	6	349 ± 25.3
	5		0.2069 gm.	6	320 ± 25.6
	6	Semesan	0.500 gm.	6	389 ± 12.9
	7		0.875 gm.	6	359 ± 24.8
	8	Sulfuric acid	4/32 fl. oz.	5	328 ± 33.3
	9		5/32 fl. oz.	5	259 ± 17.9
	10		6/32 fl. oz.	5	288 ± 12.7
	11*	Uspulun	0.500 gm.	6	403 ± 18.7
	12		0.875 gm.	6	418 ± 26.6
		Check (untreated)	.....	13	267 ± 18.7

* Treatments 4 and 11 of experiment B are equivalent in mercury content, as are also treatments 5 and 12.

sown on April 21, and those of experiment B on May 13, at the rates of 2250 and 1800 seeds, respectively. This seed was of low viability. The final stand on the aluminum-sulfate plats of experiment A show a significant increase over that of the check, while the increases with the sulfuric-acid treatments were of doubtful significance.

Unfortunately the results obtained from experiment B were somewhat vitiated by severe bird injury. As a consequence of this and of the low incidence of disease throughout the beds, the figures for final stand are significantly greater than those for the untreated plats, except for treatments 6, 11, and 12. All but treatment 9, however, gave greater absolute yields than those of the check. No chemical injury could be detected in any of the plats.

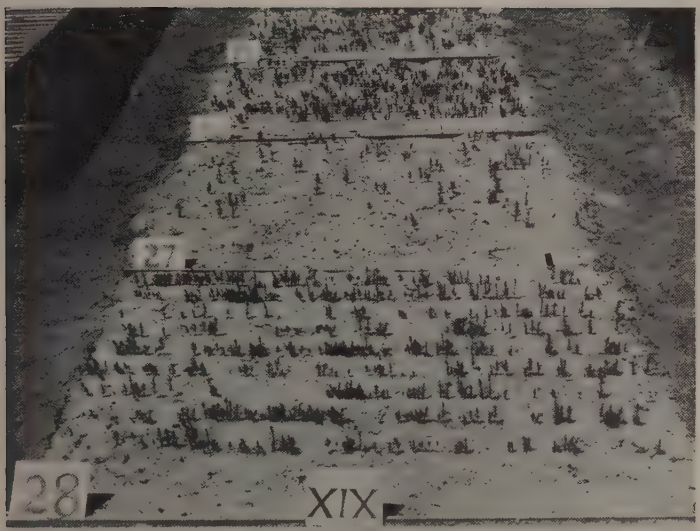
Seedlings in another experiment, in which 1 gram and 1.75 grams of Semesan and Uspulun, and 0.236 and 0.414 gram of mercuric chloride,

Plate VI

FINAL STAND OF NORWAY-SPRUCE SEEDLINGS IN BEDS TREATED WITH DRY CHEMICALS FOR CONTROL OF DAMPING-OFF

1: Plat 27, untreated; plat 25, 1/8 ounce of aluminum sulfate per square foot; plat 26, 2/8 ounce of aluminum sulfate per square foot; plat 28, 3/8 ounce of aluminum sulfate per square foot

2: Plat 33, untreated; plat 31, 4 grams of Bayer II–6 per square foot; plat 32, 3 grams of Bayer II–6 per square foot; plat 34, 2 grams of Dipdust per square foot



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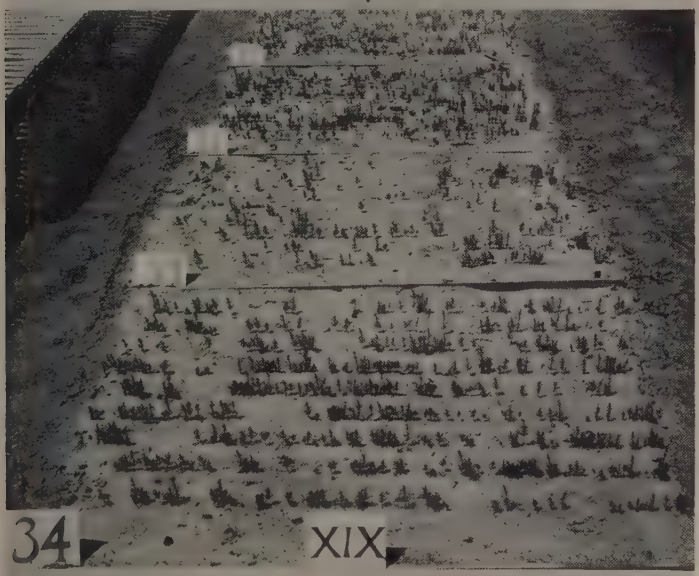

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were applied per square foot, respectively, were practically ruined through bird injury. Examination of the seedlings showed no evidence of stunting, even with the larger applications.

White pine — Treatments with dry chemicals. White pine was used, along with red pine and norway spruce, in the preliminary experiments with seed and soil applications of copper carbonate and several organic mercury dusts during the spring of 1926. The results obtained were less favorable than those with the other two species, there being lower absolute yields in some of the treated plats than in the checks.

During the spring of 1927 an extensive series of tests was conducted with applications of aluminum sulfate and a number of mercury materials. Seed of very low viability was sown on May 12, at the rate of 1800 per plat. The results of the experiment are recorded in table 18. Here

TABLE 18. TREATMENTS WITH DRY CHEMICALS ON BEDS OF WHITE PINE SOWN AT THE KEENE NURSERY ON MAY 12, 1927

Treatment no.	Treatment		Number of replications	Final stand per 9-row plat
	Material employed	Amount applied per square foot		
1	Aluminum sulfate	7.087 gms.	5	238 ± 16.5
2		10.631 gms.	5	280 ± 23.2
3		14.175 gms.	5	242 ± 21.4
4	Bayer dust	2 gms.	5	218 ± 25.0
5		4 gms.	5	275 ± 25.9
6		6 gms.	5	285 ± 9.2
7	Bayer II–6	2 gms.	5	289 ± 17.5
8		4 gms.	5	235 ± 10.3
9		6 gms.	5	274 ± 16.7
10	Dipdust	2 gms.	5	257 ± 15.8
11		4 gms.	5	248 ± 17.4
12		6 gms.	5	248 ± 10.4
13*	Mercuric chloride	0.2364 gm.	5	240 ± 15.1
14		0.4137 gm.	5	230 ± 10.0
15*	Mercurous chloride	0.2056 gm.	5	225 ± 18.2
16		0.3598 gm.	5	247 ± 16.4
17		0.3598 gm.†	5	275 ± 21.0
18	Semesan Bel	2 gms.	5	289 ± 16.0
19		4 gms.	5	265 ± 25.2
20		6 gms.	5	244 ± 5.6
21	Semesan Jr.	2 gms.	5	206 ± 18.8
22		4 gms.	5	253 ± 11.3
23		6 gms.	5	277 ± 15.0
	Check (untreated)	.....	24	153 ± 12.7

* Treatments 13 and 15 are equivalent in mercury content, as are also treatments 14, 16, and 17.
† Applied in 250 cc. water immediately after seeding.

again bird injury was so severe that detailed comparisons between different treatments cannot be made. All treatments except those numbered 4, 15, and 21, however, gave significant increases in final stand over that of the check. No evidence of stunting was apparent from any of the treatments.

Miscellaneous species. During the season of 1927, tests were made with liquid treatments of mercuric chloride, sulfuric acid, and Uspulun on beds of austrian and scotch pine, colorado, engelmann, and white spruce, and european larch. Four rows each of two different species were sown in each plat, and three replications of each treatment were made. Since the incidence of disease was low in most of the beds, the experiment was of value chiefly in determining the effects of the treatments upon the growth of seedlings during the first season. It is apparent from table 19

TABLE 19. EFFECT OF LIQUID TREATMENTS UPON AUSTRIAN AND SCOTCH PINE, COLORADO, ENGELMANN, AND WHITE SPRUCE, AND EUROPEAN LARCH

Treatment no.	Treatment		Chemical injury					
	Material employed	Amount applied per square foot in 500 cubic centimeters of water	On austrian pine	On scotch pine	On colorado spruce	On engelmann spruce	On white spruce	On european larch
1*	Mercuric chloride	0.2364 gm.	None	Slight	None	Slight to medium	Severe	Severe
2	Mercuric chloride	0.3546 gm.	None	Slight	Slight to medium	Medium to severe	Severe	Very severe
3	Sulfuric acid	3/32 fl. oz.	None	None	None	None -	Slight	Slight
4	Sulfuric acid	5/32 fl. oz.	None	None	None	Very slight	Medium	Medium
5*	Uspulun	1 gm.	None	None	None	Slight to medium	Severe	Severe
6	Uspulun	1.5 gms.	None	None	Slight to medium	Medium to severe	Severe	Very severe

* Treatments 1 and 5 are equivalent in mercury content, as are also treatments 2 and 6.

that for the most part the treatments were not injurious to the two pine species. The larger applications of mercuric chloride and Uspulun caused injury to colorado spruce, while only the acid treatments were satisfactory on engelmann spruce. European larch and white spruce were severely stunted by all the mercury treatments, while the two concentrations of sulfuric acid caused slight and medium stunting, respectively.

Effect of the treatments on germination and subsequent growth of the seedlings

In presenting the results of the soil-treatment experiments, attention was called to the injurious effects noted from the application of chemicals in certain cases. Such injury, when it resulted from treatments made at the time of seeding, was manifested by a retardation of root development

and a failure of the plant to continue normal elongation of the stem and production of leaves. Only with the copper-carbonate treatments was there evidence of that extreme type of injury expressed by an almost entire prevention of root development and by the failure of the cotyledons to grow out of the seed coats. Injury sufficient to produce killing of dormant seed was not encountered in any of these experiments. When injury resulted from a single treatment made at the time of seeding, the rate of application was in most cases higher than that necessary for effective control of the disease.

With the applications made after the seedlings had emerged, chemical injury frequently resulted, in some cases even from treatments that were ineffective in preventing the spread of the disease. This injury was manifested by both checking of root development and direct killing of stems and cotyledons.

A marked variation was found among the different species of conifers in susceptibility to chemical injury. Norway spruce was much more susceptible to such injury than were red pine and white pine. Limited tests with several other species indicated that austrian pine and scotch pine reacted similarly to red pine and white pine. Colorado spruce and engelmann spruce, however, like norway spruce, were more liable to such injury. White spruce and european larch were even more susceptible to chemical injury than were the three species of spruce noted above.

Weed control

Many investigators have reported a reduction in the number of weeds occurring in coniferous seedbeds where chemicals were applied for the control of damping-off. Similar results were noted throughout these investigations, the weeds being much fewer in most of the treated plats. The information obtained was largely observational in character, since in most of the plats weeding operations were carried out as a part of the regular nursery practice. In one experiment with colorado and engelmann spruce, however, the weeds were not removed. The condition existing in a few of the plats three months after seeding is illustrated in figure 6.

There were some indications that aluminum sulfate and sulfuric acid were in general more effective in weed control than the mercury materials, and that liquid treatments were more effective than the applications of dry chemicals.

Relative cost of different treatments

A consideration of soil treatments for controlling damping-off of conifers necessarily involves the question of the expense of making such treatments.

FIGURE 6. WEED CONTENT IN PLATS OF COLORADO AND ENGELMANN SPRUCE

The poor stand is due to low viability of the seed. The abundant spreading weed in the check plat (4) is chickweed (*Stellaria media* [L.] Cyrill.). Plat 2 was treated with 0.3546 gram of mercuric chloride, and plats 3 and 5 with 3/32 fluid ounce and 5/32 fluid ounce respectively, of sulfuric acid, per square foot

In table 20 are given the costs of representative materials which were

TABLE 20. RELATIVE COST AND RATE OF APPLICATION FOR DIFFERENT MATERIALS USED IN SOIL-TREATMENT EXPERIMENTS

Material	Type of application	Rate of application per square foot	Approximate rate of application per 1000 square feet	Approximate cost of material per 1000 square feet†
Aluminum sulfate	Dry	10.631 to 14.175 gms.	24 lbs. 7 oz. to 31 lbs. 4 oz.	$0.39 to $0.52
Aluminum sulfate	Solution	10.631 to 14.175 gms.	24 lbs. 7 oz. to 31 lbs. 4 oz.	$0.39 to $0.52
Bayer II–6	Dry	4 gms.	8 lbs. 13 oz.	$14.11
Dipdust	Dry	4 gms.	8 lbs. 13 oz.	$14.11
Semesan Bel	Dry	4 gms.	8 lbs. 13 oz.	$14.11
Mercuric chloride	Dry	0.1182 to 0.1773 gm.*	4.2 oz. to 6.3 oz.	$0.59 to $0.88
Mercuric chloride	Solution	0.1182 to 0.1773 gm.*	4.2 oz. to 6.3 oz.	$0.59 to $0.88
Mercurous chloride	Dry	0.1028 to 0.1542 gm.*	3.6 oz. to 5.4 oz.	$0.63 to $0.94
Semesan	Solution	0.50 to 0.75 gm.	1 lb. 2 oz. to 1 lb. 10 oz.	$2.87 to $4.30
Sulfuric acid	Solution	4/32 to 5/32 fl. oz.	15 lbs. to 18 lbs. 12 oz. (3.9 qts.) (4.9 qts.)	$1.05 to $1.31
Uspulun	Solution	0.50 to 0.75 gm.*	1 lb. 2 oz. to 1 lb. 10 oz.	$2.87 to $4.30

* The applications of mercuric chloride, mercurous chloride, and Uspulun noted here are equivalent in mercury content.

† Based on current prices of the grades used at the Keene nursery, shipping costs not included.

found effective in these experiments. Of the different materials tested, aluminum sulfate was the most inexpensive. The cost given is for the commercial grade, which was equally as effective as the more expensive technical grade. The organic-mercury preparations, particularly the dusts, were the most expensive of the materials used. In view of the fact that most of these have but recently appeared on the market, it is possible that the prices will be reduced with greater volume of production.

The factors of time and labor involved in applying treatments must also be considered along with the cost of chemicals. From this point of view the application of dry chemicals has much in its favor, since the inconvenience and the expense of handling large quantities of water are avoided. A further advantage of the use of dry chemicals is the ease with which the materials can be worked into the soil during the regular operations of preparing the seedbeds. The only added labor involved is that required to sift the chemical on the surface of the beds prior to the final raking.

GENERAL DISCUSSION AND CONCLUSIONS

A number of widely varying factors are necessarily involved in any discussion of damping-off. It is perhaps fortunate that in most of these investigations the causal organism was fairly constant throughout. It has been pointed out that while a number of different forms of Rhizoctonia were isolated from diseased seedlings, the majority of strains from the Keene nursery were strikingly similar. This fact is of advantage in making different soil-treatment experiments comparable to one another. One cannot, of course, draw too definite conclusions as to the results which may be expected where fungi other than Rhizoctonia are involved. Some information is available, however, from the Ithaca experiment, where Pythium was present throughout the beds and caused at least a part of the losses.

During the course of the early field experiments, considerable attention was given to the most desirable time for making soil treatments. It was hoped that some of the mercury materials would be found effective for use in checking epiphytotics of damping-off if applied after the disease first became evident. It was soon realized, however, that losses resulting from the killing of the germinating seeds and seedlings by the fungus before they appeared above ground, made such treatments impracticable.

One of the most outstanding features of these investigations has been the rather uniformly successful control of damping-off obtained through a single application at the time of seeding, with certain concentrations of nearly all of the materials tested. This is particularly striking in view of the heavy losses in the untreated plats. Thus, in many experiments the increase in final stand with most treatments over that of the check was as high as 200 to 300 per cent.

The experiments of 1927 have demonstrated that successful control of damping-off can be obtained by the use of dry applications of aluminum

sulfate, the two chlorides of mercury, and several preparations containing organic mercury compounds. As far as the writer knows, this is the first report of controlling the damping-off of conifers by this type of application. The results suggest considerable promise for the method, in point of both convenience and cost of treatments.

In using the dry chemicals at nurseries where the broadcast method of sowing is practiced, it will be necessary to disinfect the thin layer of sand which is used for covering the seed. This can be done by mixing the chemical with the sand prior to covering, or by adding a very small amount of the chemical to the surface of the beds after the seed has been covered.

Another problem arises with the use of dry chemicals, in that, with the exception of the organic-mercury preparations, none of the materials used in these tests can now be obtained on the market in the most desirable form. In the case of aluminum sulfate there is available a pulverized form from which can be sifted out a product sufficiently finely divided to be applicable for this type of treatment. It would be highly desirable, however, to have available a much more finely divided preparation of aluminum sulfate. The problem is being taken up with the manufacturers of this material.

There is also the problem of devising a suitable apparatus for applying the dry chemicals. The rather crude methods used in the present tests were effective, but they could be improved upon by employing some type of sifter or other device for obtaining a more rapid and uniform distribution of the fungicide.

With regard to the relative merits of the different chemicals, the results of these investigations indicate that liquid treatments with aluminum sulfate,[9] mercuric chloride, Semesan, Uspulun, or sulfuric acid, and with dry treatments of aluminum sulfate, Bayer II–6, or either of the two chlorides of mercury, were practically equally effective in controlling damping-off. Thus, in certain experiments significant differences could not be demonstrated between the results with these materials. In other experiments such differences existed, but the relative position of a given chemical was not constant throughout several experiments. Bayer dust, which was used only in light applications, gave less favorable results than did the above-named materials. However, since Bayer dust contains the same mercury compound (except in smaller amounts) as does Bayer II–6, it is believed that heavier applications would probably be as effective as the latter preparation. Dipdust was not uniformly so effective as the other materials. No explanation is offered, since the content of organic mercury was higher than that of other preparations which gave more favorable results. Semesan Bel, which was used to a limited extent, was effective in the larger applications. Semesan Jr. gave excellent results in several

[9] Suggested rates of application with aluminum sulfate were furnished to the writer by Dr. Carl Hartley, of the Office of Forest Pathology, United States Department of Agriculture.

tests. The results with the two last-named materials suggest that equal amounts of chlorophenol mercury are more effective when used in solution than when applied dry.

An adequate comparison of the mercury compounds on the basis of mercury content has not been made in these experiments. The data at hand, however, are suggestive in this connection. The results of the liquid treatments have indicated that mercury-equivalent solutions of mercuric chloride and chlorophenol mercury are approximately equally effective. In the dry form the two chlorides of mercury gave similar results when applications containing equal amounts of mercury were used. Nitrophenol mercury was more effective than chlorophenol mercury of equal mercury content. The results with dry applications suggest further that both compounds of inorganic mercury were more toxic to seedlings, and gave greater reductions in damping-off, than did either of the organic compounds, where the comparison is made on the basis of mercury content.

The evidence from the soil-treatment experiments definitely indicates that the materials here used exerted not only a disinfecting action at the time of their application, but a protective action as well during the period of emergence and for some time thereafter. In no instances were precautions taken to prevent reinfection of treated beds. The nursery practices made reinfection from paths easily possible, yet damping-off did not appear at the margins of the beds more frequently than elsewhere. The evidence of chemical injury from certain concentrations also indicates that the materials were present in active form some weeks after treatments were made. This is well illustrated by the fall applications on norway spruce (table 13), where injury was noted on seedlings emerging during early spring, five months after the treatments were made. Whether the chemicals remained in an active state throughout the intervening period, or whether they were brought into active form during the germination processes, is not known.

SUMMARY

This paper brings together the results of three years of investigations on the damping-off of conifers, conducted at Ithaca, New York, and at Keene, New Hampshire.

Isolations from diseased seedlings at the Ithaca nursery indicated that Rhizoctonia and Pythium were the pathogenes causing the disease. At the Keene nursery, Rhizoctonia was practically the only organism involved. Pure cultures were obtained in more than 900 cases from approximately 1300 isolations. Isolations from six other nurseries yielded a high percentage of Rhizoctonia and Fusarium, and in a few cases Alternaria.

Rhizoctonia and Pythium were highly virulent in greenhouse inoculation tests. The few tests with Fusarium and Alternaria gave negative results.

Cross-inoculation experiments with thirteen strains of Rhizoctonia were made on six species of vegetables and five species of conifers. The

results indicated considerable differences in virulence between the strains. In only a few cases did these differences seem related to the species of suscept from which the strains were isolated. Cultural characters of twenty-three strains of Rhizoctonia (including the ones noted above) are described on four artificial media. No correlation could be made between the results of the cross-inoculation studies and those of the cultural studies. With one possible exception the strains used in these tests were considered to fall within the limits of *Rhizoctonia solani* Kühn.

A limited number of observations failed to show any definite relation between damping-off and either fall seeding or application of muck to the seedbed.

The application of chemicals after emergence of the seedlings was found not to be a practicable means of controlling the disease.

Single applications of chemicals at the time of seeding, however, with any one of a number of different materials, were found to give excellent control of damping-off under conditions where extremely high losses occurred in the untreated plats. In determining the optimum rate of application, injury was noted in certain cases in which the amount of chemical used was greater than that necessary to control the disease. Austrian, red, scotch, and white pine were found to be least susceptible to such injury. Colorado, engelmann, and norway spruce were more liable to injury than the four species of pine. White spruce and european larch were severely stunted by chemical treatments in one experiment.

Applications of dry chemicals at the time of preparing the seedbeds were found to be as effective as liquid treatments.

On the basis of these experiments, it is concluded that a single treatment at the time of seeding, with liquid applications of aluminum sulfate, mercuric chloride, Semesan, sulfuric acid, or Uspulun, or with dry applications of aluminum sulfate, Bayer II 6, mercuric chloride, or mercurous chloride, are equally effective in controlling damping-off of norway spruce, red pine, and white pine. Bayer dust and Dipdust, in the amounts used, gave less satisfactory results than did the treatments noted above. Semesan Bel and Semesan Jr. were effective in the limited tests made with them. A suggested rate of application, together with the cost of materials, is presented. Aluminum sulfate is less expensive than any of the other materials. The organic mercury preparations, particularly the dusts, are the most expensive of the materials tested.

The results of these experiments indicate that at least some of the chemicals exerted not only a disinfecting action, but also a protective action for a number of weeks after the application.

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